TEXT-BOOK

OF

PHYSIOLOGY,

GENERAL, SPECIAL, AND PRACTICAL.

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REPRODUCTION,

AND

PART III.

PRACTICAL PHYSIOLOGY.

WITH FIVE PHOTO-LITHOGRAPHIC PLATES.

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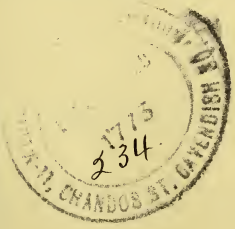

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REPRODUCTION.

The process whereby the countless variety of organisms which constitute the vegetable and animal worlds is perpetuated on the surface of the globe has from the earliest periods attracted the attention of physiologists, naturalists, and philosophers. In recent times, the excellence of the achromatic microscope has enabled us to penetrate much further into the mysteries involved in reproduction, and the whole subject is now one of vast extent.

We shall speak of this function as consisting of three kinds, viz.: first, Homogenesis; second, Parthenogenesis; and third, Heterogenesis.

HOMOGENESIS.

By Homogenesis (ὁμοιος, like; γενέσις, generation) is to be understood the production of offspring resembling in form that of their parents. This mode of reproduction is the only one found in man and the higher animals. The process may be divided into three stages: first, the production and discharge of germs; second, the fecundation of these germs; and third, the changes which follow fecundation.

The Production and Discharge of Germs.

We have seen that at the earliest period of development in all organised beings, without exception, there is formed a molecular blastema which originates a nucleated cell (pp. 45 to 49). Up to the point where sexes are manifest, the process of reproduction is identically the same with that of cell growth. The peculiarity of the function of generation in the higher organisms consists in the superaddition to this process of a particular act, whereby the further development of germ-cells is occasioned. There is a special apparatus in animals and in plants—the *ovary*,—the function of which is to mature a germ, that from the time of its first formation is capable of becoming the rudiment or embryo of a new being, and which is often separated from its parent in a form altogether dissimilar to that which it is ultimately to assume. This sometimes takes place as a spore, at others as an egg; and hence the terms *sporuliferous* and

oviparous, as distinguished from *viviparous* reproduction. The more heterogeneous a structure becomes,—that is, the more difference is manifested in the structure and properties of its separate parts,—the less title has any one to be regarded as a separate individual, since it cannot maintain an independent existence, nor reproduce the entire structure. When an organism merely consists of a multiplication of similar parts, these parts may separate, and constitute independent existences, as in the Algæ among plants, and in the Protozoa and Cœlenterata among animals. When it divides into a number of parts this has been called *fissiparous* generation—a mode of reproduction that never takes place in the more highly organised beings. In other cases, a bud is formed on the parent which may ultimately separate as an independent being. This is termed reproduction by *gemmation*. These modes of propagation are identical with that of multiplication by cells alone, with this difference, that at one period groups of cells are aggregated and united together, and afterwards separate.

Germ-cells are constantly forming and ripening in the ovaries of plants and animals, and are separated from them at particular times. In the separation of these cells, indeed, a tendency to periodicity is manifested. Thus, plants flower at certain seasons —some in spring, others in summer, and a third class in autumn or winter—with great regularity. Throughout the whole range of animals the same thing is observable. They all present a breeding period, at which time alone ova are fully developed, and capable of being fecundated.

Phenomena attending the separation of germ-cells in plants and animals.—The reproductive organs of plants and animals at this time *become elevated in temperature*. Among plants, this is most appreciable in the Arum tribe (Araceæ), where male and female flowers are collected in great numbers on a thick *spadix* or stalk, and are enclosed in a sheathing bract termed a *spathe*. On one occasion, Brogniart observed that in the *Colocasia odora* the temperature was 8° above that of the surrounding air. This was increased in the following day to 18°, and, during the emission of pollen, on the three succeeding days to 20°, after which it began to diminish with the fading of the flower.* In animals, the same elevation of temperature has caused agriculturists to denominate this season the *period of heat*. It

* "Balfour's Class-Book of Botany." 1871. Pp. 519-526.

originates in them from excessive congestion in the capillaries of the part, causing great local and more or less general disturbance of the system, the result of an augmented nutrition in the ovaries necessary for the complete development of the ova. This congestion causes rupture of the vessels and discharge of blood, which in the human female, and in a few of the monkey tribes, causes an external flow, known as the *menstrual fluid*, while the process in them has received the name of *menstruation*.

Menstruation.—This term is applied to the periodical discharge from the female generative organs of a bloody fluid. It occurs in most women once every four weeks, or once every lunar month, hence the term *menses*. It usually appears at a fixed date, and continues from three to seven days. There is then an interval of about three weeks until it again appears. The discharge is often accompanied by general symptoms, such as debility, weariness, pain in the back and limbs. It rarely occurs in pregnant women or during lactation. The quantity of fluid varies in different individuals and at different ages. The essential part of this function, however, is not the discharge of a fluid externally, but the ripening and separation of ova from the ovaries. Multitudes of seeds and of ova are formed in this manner, at regular periods, in plants and animals, which prove abortive, and the history of which is identical with the formation, ripening, and disintegration of simple nucleated cells, which have no power of reproduction.

Microscopic Characters of Menstrual Fluid. — It consists chiefly of mucus which is coagulated by acetic acid, forming molecular fibres. There are also blood corpuscles, and epithelial cells derived from the mucous membrane of the uterus. It cannot be distinguished from blood discharged from any other mucous surface, and the amount of mucus usually prevents it from spontaneously coagulating.

Structure of the Ovaries.—These organs, two in number, in the human female, are situated at the back of the broad ligament of the uterus. They measure, in the unimpregnated condition, one and a half inches in length, three quarters of an inch in breadth, and nearly half an inch in thickness. They consist essentially of a fibrous stroma (Plate XVII. fig. 4) or network, richly supplied with blood-vessels, enclosed in a tough capsule composed of white and yellow fibrous tissue (*tunica albuginea*). In the meshes of the stroma there are developed certain cells

termed Graafian vesicles, from De Graaf who first described them.* These appear first, according to Schrön† and Gröhe,‡ near the surface in the ovary of the cat (Plate XVII. fig. 4), and may be seen in great numbers in the ovary of even a newly-born female child. As they increase in size, they pass deeper into the substance of the ovary (Plate XVII. fig. 4), and undergo development, as a result of which ova are formed in their interior. In the ovary of a female at puberty, or during the child-bearing period, Graafian vesicles in all stages of development may be seen (Plate XVII. fig. 4). When the ovum is fully developed, and ready for extrusion, the cavity of the Graafian vesicle enlarges, by the secretion of fluid in its interior, pushes aside the part of the stroma between it and the surface, and projects from it externally.

Structure of the Graafian Vesicle and Ovum.—The manner in which ova are formed in the ovary has been well studied by Martin Barry, who informs us that molecules and granules are deposited in groups among the fibrous stroma of the organ (Plate XVII. fig. 4, *a*, and fig. 5). Around a large granule smaller ones are aggregated, and become surrounded by a membrane—the *ovisac*—so as to form a nucleated cell containing granular matter (Figs. 5, 6, and 7, *a*, *b*). This granular matter now separates into two portions. The inner forms a membrane that immediately surrounds the yolk, and from its transparent appearance has been called the *zona pellucida* (Fig. 4). The outer divides into two layers, one of which, covering the *zona pellucida*, he called the *tunica granulosa* (Fig. 4, *c*); and the other, which lines the ovisac, the *membrana granulosa* (Fig. 4, *b*). These two membranes are united together by four or more bands —the *retinacula*—having transparent fluid between them. In the fully formed Graafian vesicle, several of the retinacula disappear, while those remaining become shortened and enlarge so as to form a disk-shaped mass of granules, termed by Von Baer the *proligerous disk*. (See Plate XVII. fig. 4, lower Graafian vesicle.) The whole structure now forms a vesicle,—the *Graafian vesicle*,—and consists externally of a fibrous or vascular membrane, and another inner one—the *ovisac* of Barry—having suspended from it, by the *retinacula*, the ovum composed of *zona*

* De Graaf, De mulierum organis generationi inservientibus, 1672.

† Schrön, Zeitschrift f. Wissensch Zoologie, vol. xii. p. 409.

‡ Gröhe, Virchow's Archives, vol. xxvi. p. 271; xxix. p. 450.

pellucida, *yolk* (Fig. 4, *d*), and *germinal vesicle* (Fig. 4, *e*). In the interior of the germinal vesicle there is a smaller body termed the *germinal spot* (Fig. 4, *f*). So that at this period the ovum resembles a nucleated cell having also a nucleolus. Graafian vesicles, though they may be seen before puberty in the ovary, after that period increase in number and in size, and may be observed in all stages of development scattered through the substance of the organ, those most advanced being near the surface. Towards the end of each menstrual period, such as are ripe burst, from the quantity of sanguinolent serum or blood which is poured into them from the external vascular membrane, and the ovum escapes from the surface into the fimbriated extremity of the Fallopian tube, which grasps the ovary by a reflex action in order to receive it, and through which it is conveyed to the uterus. The Fallopian tube is lined by ciliated epithelium, and the play of the cilia is directed towards the uterus—in the right Fallopian tube downwards from right to left, and in the left one downwards from left to right. The ovum, however, is conveyed to the uterus, principally by the peristaltic contractions of the muscular coat of the tube.

Corpora Lutea.—The cavity thus left in the ovary is most frequently filled with coagulated blood, the result of hæmorrhage from the vascular or external layer of the Graafian vesicle, which participates in the congestion occurring in all the pelvic organs during the menstrual period. This coagulum of blood becomes gradually absorbed, in the course of which it changes its colour, and assumes a yellow and puckered appearance. The cells of the membrane granulosa multiply and grow inwards upon the clot, and assist materially in filling up the cavity.* In this state it has been called *corpus luteum* (the yellow body), (Plate XVII. fig. 9, in which one large recent *corpus luteum* is seen in the centre of the figure, an older one on the right hand, and one still older on the left.) And it has been supposed to present such peculiar appearances when fecundation has occurred as to warrant medical men in asserting that pregnancy had taken place—a grave error, which modern science has completely exploded. These appearances are described as being,—1*st*, An irregular form in the false, but a regular one in the true *corpus luteum;* 2*d*, An absence of a central cavity lined by a membrane in the false, whilst in the true there are both; 3*d*,

* Schrön and Gröhe, *ib.*

Absence of concentric radii in the false, while in the true they are present; 4*th*, The false may be present in both ovaries, while the true only exist in one. All these signs have been shewn, by numerous observations, to be in no way distinctive. Thus, in women who have never had children, there have been found *corpora lutea* exactly resembling those supposed to follow pregnancy. In the lower animals, also, four or five *corpora lutea* have been found in the ovaries, resembling each other, although one fœtus only was found in the uterus. It must be manifest that these ideas were the result of the notion that fecundation took place in the ovary, which assuredly it never does. A *corpus luteum* occurring after a pregnancy, probably disappears less rapidly than in the unimpregnated condition, but such is the only possible difference which can exist in the two states. That it is possible for any physiologist or pathologist to pronounce with certainty between the bodies which do or do not coincide with pregnancy, has been demonstrated in the negative by several remarkable cases which have been raised in courts of law. The ovaries of females advanced in life are contracted, puckered, and indurated in consequence of the numerous cicatrices that this process has produced in their texture.

Puberty.—The capability for procreation marks a peculiar period of life, which has been called puberty, on account of the development the pubes then undergoes. In woman, this generally occurs between the thirteenth and sixteenth year, but is earlier in warm climates, and later in cold ones. It has also been observed to be earlier in manufacturing towns than in thinly-peopled districts. Mental and bodily habits exercise an influence; girls accustomed to luxury and indulgence undergoing this change earlier than those reared in hardihood and self-denial. At this time those general and local changes occur which distinguish the adult woman: the mammary glands enlarge; a deposition of fat takes place in the cellular tissue of the skin, which gives to the female form its roundness and fulness; and the menstrual fluid, the most unequivocal sign of puberty, commences to flow. In man, puberty is marked by the low and rough voice—from the enlargement of the larynx by the development of the thyroid cartilage to form the *Pomum Adami*, and consequent elongation of the vocal cords; by the growth of hair on the chin, upper lips, and cheeks, as well as over the body and limbs; the greater physical power and

activity, as compared with the female ; the capability of enduring more fatigue ; and a larger amount of courage and daring. The capability of reproduction ceases in the woman, along with the function of menstruation, between the forty-fifth and fiftieth year ; but in man the term is indefinite, and virile power may continue even in very old men.

Fecundation of Germs.

The germ-cells, prepared and formed in the ovaries, are discharged from those organs at each menstrual period, and would be excreted from the economy without being further developed, unless they encountered vibratile particles formed in another organ.

Fecundation in plants.—In phanerogamous plants, the pollen falls upon the stigma, which is usually covered with a viscid matter. Minute tubes grow from the pollen, and pass downwards through the loose tissue of the style, until they reach the ovule at its base. The tube then passes through the micropyle of the ovule, and reaches the embryonal sac, and the contact of the material in the pollen tube, with the embryonal or germinal cells constitutes the real act of fecundation. Of the nature of the stimulus so imparted, we know nothing ; but the fact is well established in science that no ovule can furnish productive seeds unless the pollen has had access to it. Fecundation in many cryptogamous plants, is essentially of the same nature—the union of male and female elements produced by special organs.

Organs of fecundation in animals.—In all animals in which ova are formed the same union of male and female elements takes place. Two sets of organs analogous to those in plants are found. In some creatures, as in certain Mollusca, these are associated in one individual ; but in all the vertebrate tribes they exist in different individuals, male and female. The former is furnished with organs called the *testes*, which secrete the spermatic or seminal fluid ; the latter, with ovaries which have been already described (p. 374). The testes contain minute bodies, possessed of independent motion, which they retain for several days after they have been excreted. In them the fecundating power resides, for it is only when these come in contact with the ova discharged from the ovary of the female that the latter are ever developed into distinct living beings

From this moment that series of changes commences in the ovum whereby an embryo is formed. For this purpose, however, various circumstances are necessary, especially a fitting locality, proper temperature, moisture, &c. Seeds which have been impregnated retain the power of growth, or what some call dormant vitality, for many years; and when at length placed in favourable circumstances, they develop themselves. Generally speaking, instinct guides the lower tribes of animals to deposit their eggs in appropriate localities; and the extraordinary variety of such positions selected by insects, fishes, and reptiles, has furnished a curious subject of observation for the naturalist. In most birds, the fecundated ova are hatched by the mother, who elevates them to a proper temperature by the heat of her own body. In mammiferous animals, fecundated ova are retained in an organ—the uterus—which is provided for their reception, where they grow and become developed; and when at length they are capable of supporting an independent existence, they are excreted or parted from the body of the parent by the process of parturition.

Structure of the testes.—These organs are of an oval form, and consist of a body (Plate XVII. fig. 1, *a*, *i*, *b*,) and an elongated structure placed behind it called the *epididymis* (Fig. 1, *d*, *e*, *g*). The upper extremity of the epididymis is known as the *globus major* (Fig. 1, *d*), while the lower is the *globus minor* (Fig. 1, *g*). The gland has a tough fibrous tunic, the *tunica albuginea* (Fig. 1, *i*), which is projected inwards so as to form a prominence called the *corpus Highmorianum*, or *mediastinum testis* (Fig. 1, *c*, *f*). Numerous bands of connective tissue pass from the *corpus Highmorianum* to the capsule of the gland, thus dividing it into a number of compartments, in which lie the essential structure of the testicle, the *tubuli seminiferi*. These tubuli, originating by blind extremities at the surface of the gland, are at first much convoluted, but after passing inwards become straight, forming the *vasa recta* (Fig. 1, *b*, *s*). The *vasa recta* unite with each other in the substance of the *corpus Highmorianum*, and thus form a plexus called the *rete testis* (Fig. 1, *c*). A number of ducts, the *vasa efferentia* (Fig. 1, *d*), pass from the *rete testis*, and become convoluted, forming a series of cones, the apices of which are directed towards the *rete testis*. These cones are called the *coni vasculosi* (Fig. 1, *d*), and they constitute the chief portion of the

globus major of the epididymis. The epididymis is formed by the windings of a duct or ducts derived from the *coni vasculosi*, and at length the duct issues from the vicinity of the *globus minor*, under the name of *vas deferens* (Fig. 1, *g*, *h*). This duct passes behind the bladder, uniting with the duct of the *vesiculæ seminales*. These vesiculæ are receptacles for the storing up of semen, where it is probably mixed with mucus. The duct formed by the confluence of the *vas deferens* with the duct of the *vesiculæ seminales* is called the *common ejaculatory duct*, and opens into the prostatic portion of the urethra in a small fossa or depression, the *sinus pocularis*. The testicle is thus essentially a tubular gland. The length of the tubular structure in man has been estimated (Lauth) at 1800 feet. The appearance of one of the tubuli seminiferi, seen under a high power, is represented in Plate XVII. fig. 2. It consists of a strong basement membrane lined with epithelium, and containing molecular matter, and large cells in which the spermatozoids are developed. At certain periods few of those large cells are seen, the tubuli containing chiefly molecular matter; at other times they abound and contain one or more spermatozoids coiled up in their interior. (Plate XVII. fig. 3, *l*.)

Spermatozoids.—The form of the vibratile seminal particle varies in different animals. Various forms are shewn in Plate XVII. fig. 3 (see description of plate). In mammals generally, it has a round or oval extremity, a so-called *head*, and a filiform appendage called a *tail*, and varies in length from the ·100th to the 500th of an inch (Plate XVII. fig 3, *a* to *g*). In birds, the thick extremity is more tapering, and the whole is of a spiral form (Fig. 3, *h* to *k*). In certain reptiles and fishes, the filament is much longer, and thickest in the middle, tapering at both extremities, having occasionally a delicate continuation wound spirally round the thicker portion (Fig. 3, *m* to *r*). In some insects and crustacea, they present curious irregular forms, without a filament, and are immoveable (Lower part of Fig. 3). In the vast majority of cases, however, they possess active contractile movements. In mammals especially, when watching these under the microscope, it is difficult to divest oneself of the idea that they are animalcules, as they progress through the fluid with the heads forward, propelled by continued vibratile lashings of the tail. The notion put forth by some observers, that they possess internal organs, we have never, after careful

research, been able to confirm; and the circumstance that similar structures, with like movements, exist in the reproductive organs of many plants, negatives the idea of their being distinct animalcules. Hence, instead of *spermatozoa*, the term *spermatozoids* is more applicable to them.

Mode of fecundation.—The mode of fecundation varies in different animals. In some molluscous tribes, and in most plants, male and female organs are united in the same individual. Such an animal or plant is an *hermaphrodite*, and is usually self-impregnated. In fishes, the female sheds its spawn, and the male, swimming over it, sprinkles the spermatic fluid on the ova, and at certain seasons may be observed to follow her for that purpose. In the higher animals, union of the sexes takes place for the same end. In reptiles, especially in the frog and toad, the male clings to the back of the female, and sheds the semen over the ova immediately after they have left the cloaca. In birds and mammals, it is necessary that the spermatic fluid be deposited in the body of the female by the intromission of the male organ. From the circumstance that fecundation may take place in fishes and reptiles, as in plants, by simply sprinkling the male element over the female ova, has originated the modern practice of artificial impregnation. In the same way that horticulturists can multiply varieties, and even fertilise plants with pollen received from a distance; so, by sprinkling the fluid from the milts of male fishes over the innumerable ova which may be squeezed from the roe of the female, they may be fecundated, preserved, and reared in artificial ponds. At this moment, many of the rivers and lakes of France, Scotland, and Australia are being stored with large accessions of valuable fish so raised, in order to increase the amount of food for the people.

For a long time it was supposed that the mere contact of the vibratile spermatozoids with the ova was all that was necessary to produce fecundation; but it was first shewn by Martin Barry, and has been subsequently confirmed by many other physiologists, *that the spermatozoid actually finds its way into the ovum by a minute aperture*, so that the male and female elements ultimately blend or melt into one another. (See description of the development of the *Ascaris Mystax*, p. 48.) This fact may now be considered well established, and serves to explain many circumstances long known as to the resem-

blances which exist in feature and in qualities, mental and bodily, between parents and their offspring. Thus it has long been a matter of popular observation, that the child, in all that relates to the outward form, the gait and manners, takes after the father; while as regards the size, internal qualities and dispositions, the mother predominates. Not, however, that the male is wholly without influence on the internal organs and vital functions, or the female wholly without influence on the external organs and locomotive powers of their offspring. The law is only general, although it holds very extensively among cattle, as shewn by Mr Orton and Dr A. Harvey. Such facts seem in their turn to be accounted for by the circumstance that the spermatozoid enters and melts down in the external parts of the yolk of the egg,—that is, in connection with those layers of the germinal membrane which, as we shall subsequently see, form the nervous system and muscles; whereas the glands and internal organs are formed from the mucous layer, which is that part of the membrane furthest removed from the action of the male element.

Changes in the Ovum which follow Fecundation.

We have seen that ova are formed and discharged from the ovary at regular intervals by the adult female, but that it is only when the spermatozoid enters them that fecundation is produced. At that period the ovum presents the characters of a nucleated cell,—the *zona pellucida* being the cell-wall; the *germinal vesicle* being the nucleus; the *germinal spot* the nucleolus; while the fluid between them is opaque and granular, and called the *vitellus*, or yolk (Plate XVII. figs. 4 and 13). The size and relative amount of these three parts of each ovum vary in different animals, but they are present in all. If fecundation does not take place, the ovum degenerates, breaks down, and is ultimately excreted in the mucous discharge from the external passages. But if it encounter the spermatozoids, and one or more penetrate it, then those changes commence which terminate in the formation of an embryo. (See Plate XVII. figs. 14, 15, 18, 19, and 20, representing ova with spermatozoids in the interior.) These changes have now been followed in numerous animals, and the principal efforts of zoologists are at present directed to the elucidation of the transformations which take place at an early period in living

beings ; so that the whole subject is not only very extensive, but is constantly acquiring new facts. The study of human embryology is incomplete, for, although an ovum has been twice discovered after death in the Fallopian tube of woman (Letheby),* it has never been seen at that period when it enters the uterus. In the dog, rabbit, sheep, and other mammals, however, the various transformations have been very carefully described ; and, as it is certain that the same essential mode of development occurs in them as in man, the changes observed in the dog, according to Bischoff, will be selected as a type of what takes place in the impregnated ovum of the higher animals. (Plate XVII. figs. 12–24, and Plate XVIII.)

When the ovum leaves the Graafian vesicle, there is adherent to it externally a greater or less number of the cells which form the granular membrane. On removing these artificially, the ovum presents the appearance figured in Plate XVII. fig. 12, when magnified fifty diameters linear. It is composed of a dark, opaque yolk, surrounded by the *zona pellucida*, or vitelline membrane. On cracking this ovum between two glasses, or on tearing it with a needle, the granular yolk flows out, and the germinal vesicle escapes, as in Fig. 13, *a*. If such an ovum encounter spermatozoids, the changes subsequently represented take place. One or more enter the ovum, when they and the germinal vesicle are dissolved in the yolk,—a circumstance to which the whole structure is indebted for its continuance and for its power of, as well as direction in, development.

Development of the Embryo.

The first change observable after fecundation is that the granular yolk begins to separate into two parts,—a process accomplished by the spontaneous aggregation of the molecules of which it is composed into two masses instead of one. (See page 39 and fig. 14.) Each of these two subdivide, producing four (Fig. 15); each of these into other two (Fig. 18); and so on, until at length the whole is reduced into a mass of molecular corpuscles (Figs. 19 and 20), having a clear space or nucleus in their centres, and subsequently distinct cell-walls (Fig. 21). These corpuscles next arrange themselves in a layer externally, immediately lining the *zona pellucida*, so as to form a membrane, which is called the *germinal membrane* (Fig. 17). At one

* Philosophical Transactions. 1858.

part of this, it will be observed that the cells collect in larger numbers, and are closely packed together, forming the *germinal area*, where the embryo first appears. The ovum has now entered the uterus, and its appearance at this period, magnified ten times, is represented in Fig. 16. By cutting or tearing out the portion of the germinal membrane which contains the germinal area, and magnifying it, the subsequent changes it undergoes can be well studied. The germinal area now enlarges; at first round (Fig. 22), it becomes oval (Fig. 23), and then there appears in it a clear space,—the *area pellucida* (Fig. 24, *c*). At the same time, the germinal membrane becomes thicker, and is now divisible into two layers,—an upper or outer, called the *serous* or *animal*, from which the epidermis and cerebro-spinal system are developed; and an under or internal, called the *mucous* or *vegetative layer*, which ultimately forms the epithelium of the alimentary canal and its appendages.

The future changes in the embryo may be observed by watching the changes in these two layers, and of another that afterwards forms between them in the germinal area, called the intermediate or *vascular layer*, from which are developed all the structures between the epidermis, on the one hand, and the epithelium of the alimentary canal and appendages, on the other. In the centre of the enlarged germinal area there now forms a groove or channel, the *primitive groove*, by an elevation on each side of the serous layer of the germinal membrane (Fig. 24, *e*). This groove enlarges anteriorly, and tapers to a point posteriorly (Plate XVIII. fig. 1), and ultimately becomes closed, by its sides,—*laminæ dorsales*,—passing over it and uniting, so as to form a tube. In the floor of this tube, the embryo brain and spinal cord are differentiated in a way to be afterwards described. This tube is the cerebro-spinal canal. Underneath the canal there appears at a very early period a dense substance called the *chorda dorsalis*, a structure represented in the adult chiefly by the intervertebral discs. From the *chorda dorsalis*, as from a centre, two laminæ pass upwards,—the *dorsal laminæ*, already referred to; while two, the *ventral laminæ*, pass downwards, and meeting below, complete the body of the embryo (Figs. 1, 3, and 4). A linear mass of square-shaped cells forms on each side of the *chorda dorsalis*, the so-called *primitive vertebræ*, from which are formed the vertebral column and certain other parts (Plate XVIII. figs. 3 and 4).

The embryo is now raised prominently upwards above the serous layer (Fig. 2, which shews a lateral view of the embryo), and between it and the mucous layer another mass of cells is formed which constitutes the third or *vascular layer*, above described. Here blood-vessels are developed from large triangular cells, so as to form a plexus (Plate XVIII. figs. 3 and 4) which unites with the embryo heart and aorta (Figs. 5 and 6). Thus a circulation is established, extending over the entire ovum, with the exception of its two poles (Fig. 7). The embryo is now raised still further above the surface of the germinal membrane, while the duplications and re-duplications of its three layers, which are constantly receiving thickness by cell growth, gradually produce the various organs and textures of the body. Three vesicles or sacs are formed in connection with these layers,—the *amnios*, or amnion (from ἀμνός, a sheep, because first observed in that animal), with the serous ; the *allantois* (ἀλλάς, αλλᾶντος, a sausage ; εἶδος, shape) with the vascular ; and the *umbilical* (*umbilicus*, the navel) with the mucous layer.

The upper or serous layer of the germinal membrane may be observed from an early period to be reflected backwards, from before backwards, and laterally, so as gradually to inclose the embryo in a sac (Figs. 3 and 5). This reflexion is at first double, but after it closes over the back of the embryo, the two layers separate from each other, the outer passing outwards to be incorporated with the *zona pellucida*, while the inner forms a sac, in which the embryo is suspended. (Fig. 7). It is the *amnios* or *amniotic sac*. From the lower portion of the abdominal groove, and at the inferior extremity of the embryo, a swelling may now be observed (Figs. 10, 11, *bb*). This rapidly enlarges, and, at first open in the middle (Fig. 12, *a*), coalesces to form another sac, which hangs out of the lower portion of the abdominal opening. It is the *allantois*, a sac communicating posteriorly with the alimentary canal and the ducts of the Wolffian bodies or primitive kidneys, the ureters, Fallopian tubes and *vasa deferentia*. This sac is seen at the lower part of fig. 10. About the same time the middle layer of the germinal membrane, the vascular, splits into two layers, the inner one of which, coalescing with the mucous layer, forms the alimentary canal, while the outer is differentiated so as to form the muscles of the trunk and abdomen. The space left by the divergence of these layers of

the vascular membrane is represented in the adult by the pleuro-peritoneal cavity. As the ventral laminæ, already described, meet in the middle line of the abdomen, part of the inferior or mucous layer becomes more or less constricted by the closure of the laminæ so as to form a third sac or vesicle called the *umbilical sac.* This sac is therefore the portion of the mucous layer left outside the body of the embryo—the portion within the cavity of the embryo becoming the alimentary canal. The outer (umbilical sac) is connected with the internal part by a pedicle or duct. The mode of formation and relation of these three sacs will be better understood from the diagram seen in Plate XVIII. fig. 9, in which *a* is the back of the embryo ; *b* the amnios ; *c* the umbilical vesicle connected with the embryo by a pedicle, *d ;* and *e* is the allantois growing backwards and downwards, and continuous with the vascular layer by a pedicle *f.*

The functions of these sacs may be briefly stated to be as follows. The umbilical vesicle, containing part of the yolk, is for the nourishment of the early fœtus. The allantois brings the blood of the fœtus into relation with the surrounding media for the purposes of nutrition and respiration. It is seldom of large size in mammalia, because it is soon supplanted by another organ, *the placenta,* into the structure of which it enters. The amnios secretes a fluid, termed amniotic fluid, in which the fœtus floats, and by its reflexions it permits the *allantois* to pass outwards so as to contribute to the formation of the placenta.

Development of the Chorion.—While the ovum is very small, its outer surface becomes shaggy from the appearance of numerous villi. These villi, at first simple, soon become branched by lateral processes passing from them (Plate XVIII. fig. 11). This villous covering is the *chorion.*

*Development of Special Organs in the Embryo.**

Development of the skeleton.—Immediately below the primitive cerebro-spinal canal there appears in the middle or vascular layer, a structure termed the *chorda dorsalis,* or notochord, consisting of large cells, surrounded by a thin sheath. About the same time, small square-shaped masses are to be seen on each side of the *chorda dorsalis* (Plate XVIII. figs. 1, 2). These are the *primordial vertebræ,* and each pair is ultimately

* For further details reference is made to works specially devoted to the subject. See also description of Plates XVII. and XVIII.

developed by differentiation into the osseous and cartilaginous portions of a permanent vertebræ, the head of a rib, the central parts of a spinal system of nerves, and the cutaneous and muscular parts covering the back.* During this process of development, the *chorda dorsalis* becomes constricted, and ultimately portions of it form the centres of the bodies of the permanent vertebræ, while other portions are persistent in the intervertebral disks. Thus the vertebral column is formed.

Development of the skull.—At a very early period of fœtal life, two curvatures are to be observed near the anterior extremity of the embryo, one at the point corresponding to the junction of the vertebral column with the skull, and the other opposite to the second cerebral vesicle. Behind the latter curvature, the dorsal plates bend downwards and unite inferiorly, so as to form four arches, behind each of which there is a cleft or fissure, termed a *branchial cleft* (Plate XVIII. fig. 14, *d*, *f*). The posterior part of the first cleft remains open in the fully developed fœtus as the external aperture of the ear, the cavity of the tympanum, and the Eustachian tube, while its anterior part, along with all the other clefts, are ultimately obliterated. We have now to consider, first, the development of the cranium, and second, that of the face.

1. *The cranium.*—The *chorda dorsalis* terminates at the posterior part of the *sella turcica*, the fossa in the base of the skull for the reception of the pituitary body. In front of this point, two thick bars of cartilage, separated by a thinner portion between them, pass forwards, and unite in front of the ethmoidal region. These have been termed the *lateral trabeculæ* of Rathke. The whole of the base of the skull is now cartilaginous, while the vault of the cranium is membranous. The cartilaginous portion ossifies and differentiates into the occipital bone below its protuberance, the petrous and mastoid portions of the temporal, the sphenoid and ethmoid; while the membranous portion becomes the parietal, frontal, upper part of the occipital, and the squamous portion of the temporal. The vomer and perpendicular plate of the ethmoid are developed from a vertical process of cartilage passing forward from the neighbourhood of

* Quain's "Elements of Anatomy," 1867, p. 16; Goodsir's "Anatomical Memoirs. On the Morphological Constitution of the Skeleton of the Vertebrate Head," vol. ii., p. 89, *et seq.*

the termination of the chorda dorsalis, called the *ethmo-vomerine* cartilage.*

2. *The face.*—The bones of the face are all originally cartilaginous, and are developed as follows: A process passes from the upper or first visceral arch forwards beneath the eye, and forms the sides of the face, namely, the superior maxillary and malar bone (Plate XVIII. fig. 14, *e*). Coincident with the development of the two processes just described, another process, called the *middle frontal process*, passes down from the anterior extremity of the ethmo-vomerine cartilage between them, and becomes the nose and middle part of the upper lip. The lower part of this process bears the upper incisor teeth. In man it is blended, even at a very early period of fœtal existence, with the *superior maxillary* process; but in all other mammals it remains separate as the *intermaxillary* or *premaxillary* bones. In the three upper visceral arches, narrow strips of cartilaginous tissue make their appearance. These are the subjects of remarkable changes. The first, or upper piece, is divided into three parts: the proximal, or that in connection with the *basis cranii*, is developed into the palate plate and internal pterygoid process of the sphenoid bone; the second forms the incus and its two processes; while the remaining part, long and narrow, passes downwards and forwards so as to unite with its fellow of the opposite side, and is called *Meckel's cartilage.* The upper part of this cartilage forms the malleus and its handle, and the lower part forms a rod, on the external surface of which the lower jaw is developed. It ultimately disappears, except a small portion, which is represented by the *processus gracilis* of the malleus. The proximal extremity of the firm tissue in the second arch forms the stapes, and the distal extremity the styloid process of the temporal, the stylo-hyoid ligament, and the small cornua of the hyoid bone. A portion of that of the third arch forms the great cornua and body of the hyoid bone. Thus are formed the bones of the face and ear.

Development of the limbs.—The upper and lower limbs are developed from the ventral plates on the sides of the body. The upper limb appears before the lower, and according to Kölliker, the division into arm and fore arm, thigh and leg, occurs about the eighth week. About the same time the division

* Quain's "Elements of Anatomy," 1867, p. 65.

into fingers and toes also takes place (Plate XVIII. figs. 10, 13, 15).

Development of the organs of circulation.—The heart is at first a mass of cells, shewing rythmical contractions even before muscular tissue is developed, but at an early period it is a simple dilated tube having two veins entering its posterior end and a large arterial trunk passing from its anterior. The tube soon becomes constricted in two places, so as to form three compartments, the posterior of which is the auricular portion, the middle the ventricular, and the anterior the *bulbus arteriosus.* Septæ next make their appearance in these compartments, so as to divide the auricular portion into the two auricles, the ventricular into the two ventricles, and the bulbus portion into the aorta and pulmonary artery. The heart also becomes twisted upon itself somewhat like the letter S, and this process goes on until the two auricles are anterior, at the base of the heart, the two ventricles posterior, forming the cone. From these originate the pulmonary artery and the aorta (Plate XVIII. figs. 3 and 6). The bulbus arteriosus is at first a tube which, after passing forwards a short distance, splits into two. These diverge, but afterwards unite to form a large tube running down behind the heart in front of the vertebral column. This tube is the descending aorta. Thus two arches are formed. Four other pairs of arches are also developed, each arch being placed in one of the branchial processes already described (fig. 14). These arches never co-exist, as the highest disappear before the lower are developed. This transitory arrangement of blood-vessels resembles somewhat the branchial arteries in fishes; but it soon disappears. Embryologists have not satisfactorily traced the development of these vascular arches into permanent structures. According to some, the fifth or uppermost arch remains persistent as the anastomosis between the internal carotid and vertebral arteries through the circle of the Willis at the base of the brain; the fourth, as the inosculation between the superior thyroids of the external carotids and the inferior thyroids of the subclavian; the third, as the subclavian arteries; the second becomes, on the left side, the arch of the aorta, while that on the right side disappears; and the first is represented in the fœtus by the *ductus arteriosus* on the left side, the right having disappeared at an earlier period.*

* For further details regarding the development of the great arteries and veins,

represented in the adult by the floor of the fourth ventricle, communicating below with the canal of the spinal cord. The anterior part of the roof is differentiated into the cerebellum, and the transverse commissural fibres of the cerebellum constitute the *pons Varolii*. The *vermiform process* of the cerebellum appears before the two lateral hemispheres. In the floor of the middle or second cerebral vesicle, matter is deposited so as to form the *crura cerebri*, and in the roof we have developed the *corpora quadrigemena*, an antero-posterior median groove being first seen about the sixth month, and a transverse, separating the *testes* from the *nates*, first making its appearance about the seventh month of intra-uterine life. The primitive cavity of the second cerebral vesicle remains persistent as the Sylvian aqueduct, or *iter a tertio ad quartum ventriculum*. The development of the anterior primary cerebral vesicle is more complicated. At a very early period, two vesicles are developed from the anterior primary vesicle, one on each side. These vesicles have been termed the *hemisphere vesicles*, because from them are developed the hemispheres—the *corpora striata* appearing in the floor, while the hemispheres, properly so called, constitute the roof. The external surface of the mass termed *corpora striata* is the *Island of Reil*, seen in the Sylvian fissure. The cavity of these vesicles is represented by the lateral ventricles, and between the double partition separating them, we have the fifth ventricle. The cavity of the anterior primary vesicle, behind the cerebral vesicles, forms the third ventricle, the floor being formed by the *optic thalami* which are at first hollow, while the roof is formed by the *velum interpositum*, a layer of *pia mater*, which is folded into the brain through the transverse fissure. As development proceeds, the communication between the cavity of the anterior cerebral vesicle (third ventricle) and those of the hemisphere vesicles (lateral ventricles) becomes smaller and smaller, and ultimately constitutes the *foramen of Munro*. The margin of this foramen forms anteriorly the *fornix*, an antero-posterior commissural set of fibres, and posteriorly the *corpus fimbriatum* and *hippocampus major*, and as the hemispheres increase in size they grow backwards, so as to overlap the *optic thalami*, *corpora quadrigemena*, and cerebellum. The great transverse commissural mass, *corpus callosum*, is first seen about the end of the third month. The first trace of convolutions is seen about the fourth month. They are at first indistinct, and continue so till

the seventh month, after which they are rapidly developed. The Sylvian fissure appears at the fourth month, and is soon followed by the fissure of Rolando. Such is an outline of the development of the brain.

Development of the eye.—The eye first appears as a hollow process in connection with the anterior cerebral vesicle (Plate XVIII. fig. 5, where the first cerebral vesicle is expanded laterally; fig. 6; fig. 8, *e*; and fig. 13, *b*). This process soon becomes a round vesicle (*primary optic vesicle*) connected posteriorly with the anterior cerebral vesicle by a hollow pedicle. The optic vesicle now approaches the cuticle and becomes invaginated, carrying a portion of the cuticle along with it. This invaginated portion of cuticle, at first a pouch, becomes constricted to form a sac, and is ultimately severed from its connection with the general cuticle, thus forming the *lens*. The primary optic vesicle now undergoes a second invagination behind the lens until the opposite surfaces come into contact, and the cavity of the primary optic vesicle disappears. According to Kölliker, the invaginated portion forms the retina and the layer of hexagonal pigment cells in the choroid; and the outer portion, the pigmentary (branching pigment cells) and probably the vascular part of the choroid. The cup-shaped cavity behind the lens, called the *secondary optic vesicle*, is soon filled with the vitreous humour. The iris is developed about the second month as a septum projecting from the anterior part of the choroid. The sclerotic and cornea are formed from tissue external to the eye. The lens in the fœtus is surrounded by a vascular tunic, the fore part of which is persistent in the young of many animals for several days after birth. It is then termed the *pupillary membrane*. In the human being it is atrophied before birth. The eyelids are folds of integument. The lachrymal canal is the representative of the fissure between the frontal process and the maxillary lobe of the embryo.*

Development of the ear.—The ear is developed from a vesicle, the *primary auditory vesicle*, above and behind the second branchial arch (Plate XVIII. fig. 5, where the embryo ears are seen opposite the third cerebral vesicle; fig. 8, *f*). This vesicle, however, is not a process from the brain, as is the case in the eye, but an invagination of the cuticle. The first part developed is the membranous labrynth, afterwards the semicircular canals,

* Quain, vol. ii. p. 737-9.

and finally the cochlea. The osseous labrynth is developed from cartilage, continuous with the cartilage at the base of the primordial cranium. The stages of development of the intricate portions of the cochlea have not yet been clearly made out. The development of the middle and external ear have been already described. The external meatus, tympanum, and Eustachian tube are the remains of the upper part of the first branchial cleft ; the incus and malleus are formed from the upper part of Meckel's cartilage (part of the first branchial arch), while the stapes is derived from the cartilage of the second branchial arch.

Development of the nose.—The nose is a development of the integument (Plate XVIII. fig. 13, *a*). The olfactory bulbs, at first hollow, are processes from the two vesicles which ultimately form the cerebral hemispheres (p. 394). It has not yet been made out whether the olfactory nerves grow from the bulbs, or originate, like other nerves, from separate masses of blastema. The nostrils first appear as two grooves separated by the frontal process. They are shut off from the eye by the lateral frontal process, and the side wall of the nostril is completed by the maxillary processes. The nostrils at this period communicate with the mouth. The palate is now developed by transverse growths towards the middle line, and ultimately they unite. Sometimes we have, by their non-union, the congenital deficiency known as cleft-palate.

Development of the alimentary canal.—The alimentary canal first appears as a groove directed towards the yolk. The groove is lined by the mucous layer of the embryo, which ultimately forms, as already mentioned (p. 384), the epithelium of the canal. The wall of the groove is formed by the deeper division of the intermediate or vascular layer of the embryo. The groove soon closes, so as to form a long, straight tube, stretching in front of the vertebral column from the base of the skull to near the posterior extremity of the embryo (Plate XVIII. fig. 13, *m*, and fig. 15, *l*). This tube, however, communicates with the yolk-sac, or umbilical vesicle, by an opening on the ventral aspect, which speedily becomes a narrow duct, named the *omphalo-enteric duct* (Fig. 9, *d* ; fig. 15, *m*). This duct soon disappears. The intestine now forms a curve or loop in the centre of the body, and a portion dilates to become the stomach (Fig. 13, *e*, and fig. 15, *k*). The stomach is at first vertical, turns

over on its right side, so that the right lateral surface becomes its posterior surface. This explains the anatomical fact of the right pneumogastric supplying the posterior aspect of the stomach, while the left supplies its anterior aspect. The great intestine, at first narrower than the small, in the early embryo shews no cæcum. The mouth is developed by an infolding of the integument above the highest branchial arch, and is separated at first from the pharynx by a partition. The anus and lower part of the rectum are also invaginations of the outer surface. It is remarkable that villous processes are at first found throughout the whole alimentary canal, but ultimately disappear in the stomach and large intestine. Occasionally, diverticula are found in the adult in connection with the lower part of the ileum. These are believed to be remains or developments of the original omphalo-enteric duct. Umbilical hernia arises from the want of complete apposition of the wall of the abdomen at the umbilicus.

Development of the liver.—The liver appears as two cul-de-sacs or tubes, arising from the intestinal canal immediately beneath the dilation for the stomach (Plate XVIII. fig. 13, *k*, and fig. 15, *i*, *i*, *i*). These processes, according to Remak, consist both of the epithelial and muscular parts of the intestine. They increase in size by the development of glandular substance, and surround the *omphalo-mesteric vein*, the vein bringing blood from the umbilical vesicle, and from the wall of the primitive alimentary canal (Figs. 6 and 7). As the umbilical vesicle decreases in size, the omphalic portion of the vein also decreases, and the mesenteric portion coming from the alimentary canal becomes the *portal vein.* The continuation of the omphalo-mesenteric vein passing from the liver to the general circulation receives the name of the *hepatic vein.* At this period, then, the liver is supplied with blood chiefly by the portal vein. Coincidently with these changes, the allantois is being developed so as to form, along with the decidual membranes, the placenta ; and as the placenta increases in size, a time arrives when the liver receives more blood by the *umbilical vein*, or vein coming from the placenta, than from the portal vein.* The umbilical vein, on reaching the liver, divides into two branches ; one of these, the smaller, passes onwards to the *vena cava inferior*, and is called the *ductus venosus ;* the other joins the *vena portæ.*

* Dalton's Physiology, p. 664.

Several other small branches are given off by the umbilical vein to the liver. After respiration is established, and blood ceases to come by the umbilical vein, it and the *ductus venosus* contract and shrivel up, the umbilical vein constituting the round ligament of the liver, while the *ductus venosus* disappears. (See pp. 224, 225, and Plate XI. fig. 16, and description.)

Development of the salivary glands, and pancreas.—These organs are developed as simple canals with small processes passing them. In the case of the salivary glands, these canals communicate with the mouth, while that of the pancreas arises from the left side of the intestine, close to the spleen.

Development of the organs of respiration and organ of voice.—The lungs are at first diverticula from the œsophageal portion of the alimentary canal, and their internal cavities are lined by a prolongation of the lining of the œsophagus. At a later period, they are connected with the digestive tube by a pedicle which becomes the trachea (Plate XVIII. fig. 15, *a*). The lungs are seen at a very early period of development, and the air cells are rapidly developed round the extremities of the ramified bronchial tubes. Until birth, the lungs are of small size, and occupy a small space at the back of the thorax. On respiration being established, they expand so as to fill the cavity. The rudimentary larynx appears as two slight enlargements separated by a fissure, and embracing the communication between the pharynx and trachea. According to Rathke, all the true cartilages are formed at the same time. The larynx is small in childhood. In the female, the larynx retains its comparatively small size, and rounded thryroid cartilage anteriorly ; but in the male the cartilages become stronger, and the alæ of the thyroid cartilage project forwards so as to form the *pomum Adami*. The vocal cords are thus lengthened. The cartilages undergo partial ossification from middle life to old age.

Development of the blood glands.—The *spleen* appears about the seventh week, close to the pancreas, but by the tenth week it is placed at the great end of the stomach. It is developed in a special mass of blastema. Kölliker has observed the *thyroid gland* at the end of the third month, as consisting of shut sacs with cells in their interior. This organ is relatively larger in the fœtus than in after life. The *thymus gland* (Plate XVIII. fig. 15, *b*) has been seen by Simon in embryoes of swine and oxen about half an inch in length, as a simple tube. From

this tube lateral diverticula arise containing corpuscles. By the twelfth week, the thymus consists of lobules; it rapidly increases in size during fœtal life, and continues to grow after birth to the end of the second year. By the tenth or twelfth year it becomes a fatty mass, and at puberty it has almost disappeared. The *supra renal capsules* originate from blastema different from that of the kidneys. Some observers (Goodsir) have seen at an early period these organs apparently in one mass, others in two separate masses united together, while many have seen them at birth completely separated from each other. At one period, about the fifth or sixth week, the supra-renal capsules are larger than the kidneys, but ultimately they become much smaller. Nothing whatever is known definitely of the development of the lymphatic glands, Peyer's glands, pituitary body, or pineal gland.

Development of the urinary and generative organs.—At a very early period in the development of the human embryo, two ridges of blastema appear, one on each side of the primitive straight alimentary canal. These increase in size,—vesicles, and afterwards tortuous cæcal tubes, are developed in them, communicating with a duct which runs along the outer side of each organ. These are the *primordial kidneys* or *Wolffian bodies*, named after their discoverer, C. F. Wolff (Plate XVIII. fig. 13, *o*; fig. 15, *o*). The ducts open into the allantois, and a whitish excretion, containing uric acid, has been found in them and in the allantois. As development progresses these bodies decrease in size, and ultimately almost entirely disappear, their ducts, however, becoming modified into certain permanent structures, as will be afterwards described.

The *kidneys* are not developed from the Wolffian bodies, but from separate masses of blastema behind their upper end. At first smooth, the kidneys soon become lobulated, each lobule consisting of a number of cæcal tubes which are modified into the *tubuli uriniferi*. The *Malpighian bodies* have been seen at a very early period. The *ureters* are believed by Rathke to commence separately from the kidneys, and afterward to become united to them. The ureters open into the allantoid sac. The lower part of the allantois is retained in the abdomen by the closure of the ventral walls at the umbilicus, and receives the ducts of the Wolffian bodies and of the ureters. This portion becomes dilated, and afterwards constitutes the *urinary bladder* (Fig. 9, *f*). The contracted part above this dilated portion,

passing from the bladder to the umbilicus, is afterwards known as the *urachus*. The bladder at first communicates freely with the lower end of the alimentary tube. This general cavity is called the *cloaca*, and represents the permanent condition in birds and reptiles. Soon, however, a partition is developed, dividing the cloaca into two parts, the posterior constituting the *rectum*, opening externally by the anus, while the anterior has been termed the *sinus urogenitalis*, and receives the ducts of the Wolffian bodies as well as those of the kidneys and of the ovaries or testes.

The *generative organs* are developed from separate masses of blastema on the inner aspect of the Wolffian bodies. It cannot at first be determined whether male or female organs are to be the result of the development. The Wolffian bodies are pushed outwards by the development of the kidneys above and between them, and they gradually decrease in size. A white elongated thread-like mass of blastema is seen on the front of each Wolffian body, running along the duct. This becomes a tube termed the *Müllerian duct*. The Müllerian ducts become fused together at their lower end, and, along with the ducts of the Wolffian body, form a single cord—the *genital cord*. Now commences a distinct differentiation between the organs of the sexes. In the female, the portion of the Müllerian ducts united together is developed into the *vagina*, *cervix*, and lower part of the *body* of the *uterus*. This partial formation of the uterus by the union of two ducts explains the occurrence occasionally of a double or horned uterus. The upper part of the body of the *uterus* and the *Fallopian tubes* are formed by the remaining upper portions of the Müllerian ducts. A very different development occurs in the male. Here the Müllerian ducts undergo little development, and are represented in the adult by the *vesicula prostatica*, *sinus pocularis*, or *utricle*, first described by Morgagni, and situated in the floor of the prostatic portion of the urethra, and in front of the *caput gallinaginis*, or *verumontanum*. This little recess, therefore, represents in the male a portion of the uterus and the vagina in the female. The united portions of Müller's ducts disappear. The male organs of generation are developed partly from the mass of blastema already referred to, and partly from the ducts of the Wolffian bodies. The ducts of the latter form the *vasa deferentia* and *ejaculatory ducts*, the *vesicula seminales* being cul-de-sacs from their lower part. Part of the duct of the

Wolffian body becomes the *epididymis.* According to Cleland and Banks the *coni vasculosi* are developed from separate deposits of blastema. The testicles are, until the seventh month, in the abdominal cavity. They then descend through the internal inguinal ring into the scrotum pushing a pouch of peritoneum before them. This pouch becomes constricted, and is ultimately cut off from the abdominal portion of the peritoneum, so as to form the *tunica vaginalis.* The external organs of generation are first represented by a small body projecting from the median line in front of the cloacal opening, having a groove on its under surface. This body is the rudimentary organ which becomes the *clitoris* in the female and the *penis* in the male. About the eleventh or twelfth week, a transverse band divides the anal from the genito-urinary passage. Two cutaneous folds, one on each side of the clitoris, become, in the female, the *labia majora.* In the male, the penis enlarges, and the margins of the groove on its under surface unite so as to form the *urethra.* The lateral cutaneous folds already mentioned unite so as to form the scrotum. When the union of the urethra canal is not complete, and the penis is small, we have then the condition of hypospadias.

In the human ovum similar changes occur to those in the dog. The early stages, indeed, may be said to be identical. The umbilical vesicle and allantois, however, never become so large, and according to a dissection of a fœtus by Müller, supposed to be twenty-eight days old, union between the two may be observed even at that early period. The amnion is formed in the same manner as in the dog, very early, and continues during the whole period of intra-uterine life, constituting the membrane that surrounds the fœtus. On the chorion, the villi become concentrated on a particular spot, while the rest of its surface becomes smooth. The allantois grows into this part of the chorion, and blends with it, as will be afterwards more particularly described. About the beginning of the second month, the umbilical vesicle begins to disappear, it shrinks together, its peduncle uniting with that of the allantois to constitute the umbilical cord. Its remains may sometimes be traced in the fœtal coverings at birth. All the organs are now evolved, and are gradually perfected during the remainder of intra-uterine life.

In birds. — The same process previously described occurs essentially in the egg of the bird. In the bird there is no uterus. The ovum is maturated out of the body of the animal. In the ovary of the common fowl, numerous eggs may be seen in various stages of development. When discharged from the ovary into the Fallopian tube or *oviduct,* the egg consists of a large globular yellowish yolk inclosed in a transparent vitelline membrane. At one spot on the surface of the vitellus, there is a round white spot, the "*cicatricula,*" in the centre of which is the germinal vesicle. The peristaltic actions of the muscular wall of the oviduct forces the egg downwards, and during its passage a homogenous layer of a gelatinous deposit is formed around the vitelline membrane. This is termed the "*chalaziferous membrane.* The egg is still pushed downwards with a slow rotatory motion, and the chalaziferous membrane is twisted in opposite directions so as to give rise to two fine cords, running from the opposite poles of the egg, termed the "*chalazæ ;*" and from the mucous membrane of the oviduct there exudes an albuminous substance, the so-called albumin, or "*white of egg.*" In the next part of the oviduct, another material is deposited outside the albumin, in the form of three distinct membranes, layers, or envelopes ; and in the lower part of the oviduct, which is wider than the rest of the canal, the villous mucous membrane transudes a fluid containing a large amount of calcareous salts, which are deposited amongst the fibres of the outermost layer. Thus, the shell of the egg is formed. The egg is finally discharged through a narrow portion of the oviduct, and finally from the external orifice.

The shell, although it has no visible pores, is still permeable to air, a condition essential to the future development of the embryo, for it has been found that if the shell be covered with a layer of varnish, the egg dies. The ovum of the bird contains all that is necessary to perfect the embryo, and it receives no further nourishment from the mother. On this account it is furnished with a much larger amount of albumin and yolk, which are metamorphosed by cell growth, into the tissues of the chick. During this metamorphosis some chemical change is continually going on, and material, such as watery vapour, must pass out through the shell, for the egg is daily losing weight. Soon after the egg is passed, a small quantity of air passes through the shell at the rounded extremity, and accumu-

face—the portion opposite to the allantois. This is the situation of the future placenta. This organ consists essentially of a fœtal and a maternal portion. The *fœtal* part is formed by the villous tufts of the chorion, containing loops of capillaries derived from the allantois. It is connected with the fœtus by the umbilical cord, and both it and the cord are covered by a reflection of the amnion. The *maternal* portion consists of the decidua, or hypertrophied mucous membrane, containing the uterine follicles. As already explained, these follicles become much enlarged, so as to allow the tufts of the chorion to be pushed into them, and at the same time the blood-vessels or veins ramifying on the outer surface of the follicles rapidly increase in size, and thus form large sinuses, termed the *uterine sinuses.* These sinuses exist in the decidua, not in the wall of the uterus, and they and the uterine follicles mould themselves over the outer surface of the villous tufts of the chorion, as seen in Plate XVII. fig. 8, *a*, *b*, *c*, *d*. Thus the blood of the fœtus is in close proximity to that of the mother, but is separated from it by the following membranes, which become more or less fused together: first, the wall of the fœtal capillary (Plate XVII. fig. 10, *g*); second, the chorion (*e*, *f*); third, the wall of the uterine follicle (*b*); and fourth, the wall of the uterine sinus (*a*). According to Goodsir, there are two layers of cells between the fœtal and maternal portions of the placenta,—first, a layer on the lining of the uterine sinus, *b* (*external cells*); and second, a layer on the chorion, *e*, *f* (*internal cells*). The function of the external cells is to absorb nutrient matter from the blood of the mother, while that of the internal is to absorb this matter from the external, and pass it into the blood of the child.* Through these membranes, the blood of the fœtus receives nutritious matter from, and gives up impurities to, that of the mother, probably by processes of endosmose and exosmose. During pregnancy, the involuntary contractile fibre of the uterus becomes gradually hypertrophied so as to increase the power of the uterus.

Parturition.—The process of parturition takes place a few days beyond the end of the ninth calendar month. During pregnancy, the involuntary contractile fibre of the uterus is enormously hypertrophied. The fœtus is expelled by the

* Goodsir. The Structure of the Human Placenta. "Anatomical Memoirs," p. 445.

contractile force of the uterus, aided by the abdominal and other muscles. It is soon followed by the expulsion of the placenta. When the placenta separates, the uterine vessels are torn across, but the orifices are speedily closed by the contraction of the uterus. The whole placenta, maternal and fœtal, including the uterine sinuses, uterine glands, and tufts of the chorion, separates, and the hæmorrhage which usually takes place does not come from the uterine vessels, but from the large sinuses in the placenta. After parturition, the muscular walls of the uterus atrophy by fatty degeneration, and the mucous membrane is renewed.

General conclusions.—Such is a general sketch of the various stages of the function of reproduction, a study of which in the different classes of animals has led to the formation of various ingenious hypotheses, whereby it has been sought to bring the order of evolution within the operation of certain laws. One of these, which has excited great attention, is, that the human fœtus passes through transition periods resembling in turn the different inferior beings of the animal scale : that is to say, it at first resembles a zoophyte, then a mollusc, then a worm, a fish, a reptile, and so on. Thus the monads found among the inferior animals have been supposed to be represented by the germinal vesicle. The yolk, when divided, has been thought to resemble a gonium or a volvox. When the primitive groove closes, it has been likened to a worm ; afterwards to a molluscous animal ; and when the visceral arches appear, to a fish ; and so on. This method of viewing the phases of development has led to a generalisation thus expressed by Serres,—viz., that "Human organogenesis is a transitory comparative anatomy, as in its turn comparative anatomy is a fixed and permanent state of the organogenesis of man." But that the human embryo ever resembles a worm, a mollusc, reptile, fish, or bird, can, on careful examination, nowhere be recognised. It is true that at one period all ova resemble each other ; but it is equally certain that from the first moment of their formation they are impressed with a power of developing themselves in a certain direction, so that the ovum of a reptile, fish, or bird, will always be developed into similar animals, and by no concurrence of circumstances will ever be transformed into different ones. Neither is there any anatomical or structural relation between

them, for the visceral arches in the human fœtus are in no way, as has been supposed, analogous to the branchiæ or lungs of the fish, for the former are partly transformed into the bones of the face, while lungs originate in inflections of the mucous layer. The theory, then, may be considered as more fanciful than real, and founded upon loose analogies, which, instead of being strengthened, are weakened as development proceeds, and the true types of such analogies become more evident.

Lactation.

The most natural food for the infant is the milk of the mother. This fluid is secreted by special glands termed the *mammary glands.* In the human female, they form two rounded masses, the breasts, placed one at each side on the front of the thorax. They are essentially compound racemose glands, resembling in structure the pancreas and salivary glands. They consist of a number of lobes or lobules, from which proceed about fifteen or twenty ducts, called *galactophorous ducts.* These converge towards the areola, or circle round the prominence, or nipple, where they form *sinuses.* From these sinuses, small ducts pass to the surface of the nipple, where they open by separate orifices. When examined by a high power, the small vesicles and ducts of the gland are found to consist of a wall of areolar tissue lined by a mucous membrane, having tasselated epithelium. They are usually filled with molecular matter. The secretion of milk commences during the latter period of pregnancy, but it is not fairly established until two or three days after delivery. Its appearance is usually ushered in by a feverish condition, "*milk fever.*" The fluid secreted during the first few days after delivery is of a yellower colour than that secreted afterwards, and is termed *colostrum.* This milk is believed by some to act as a natural purgative to the infant. Afterwards the milk is white, or bluish white, opaque, has little or no smell, and a slightly sweet taste.

Histological structure of milk.—When a drop of milk is examined with a magnifying power of 250 diameters, it is found to consist of a fluid in which there are numerous globules of various sizes, varying from the 1/2500 to 1/1500 of an inch in diameter (Fig. 1, p. 404). These globules are very refractive.

They are often in groups, but in healthy milk readily roll upon one another. On the addition of acetic acid they may be seen melting together, so as to form larger globules, which may then be dissolved by ether. Thus we learn that each milk globule consists of an envelope of albumen surrounding a drop of oil, and an appearance very similar to milk can be artificially prepared by first forming the *Haptogen membrane of Ascherson*, as already described at p. 36, and then rubbing the covering glass with a circular motion, so as to break up the membrane. Colostrum is found to contain, in addition to the ordinary milk globules, numerous large, irregular, globular bodies, from the 1/1100 to 1/800 of an inch, termed "*colostrum corpuscles.*" They contain numerous minute oily granules (Fig. 2).

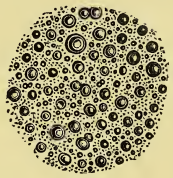


Fig. 1.

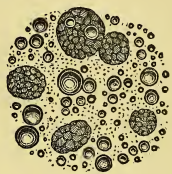


Fig. 2.

Chemical composition of milk.—The specific gravity of human milk is about 1032 (*Simon*). It is usually alkaline, sometimes neutral, but it gradually becomes acid. When it has stood for a time, the globules float to the surface forming a layer called *cream*, while beneath we have a bluish-white fluid, poorer in fat, but having a higher specific gravity than the upper stratum. If the temperature is cool, and the atmosphere not highly charged with electricity, milk will remain in its natural condition for several days; but under reverse conditions, it spontaneously coagulates. This coagulation is caused by part of the milk sugar undergoing acid fermentation, yielding lactic acid, which precipitates the albuminous constituent of milk—the casein.

$$\underset{\text{Milk Sugar.}}{C_{12}H_{20}O_{10}} + \underset{\text{Water.}}{2H_2O} = \underset{\text{Lactic Acid.}}{4(C_3H_6O_3)}$$

The following are analyses of human milk, compared with that of a few other animals, in 1000 parts :*

* Article "Milk," in Watt's Dictionary of Chemistry, vol. iii.

Constituents.	Human—*Vernois* and *Becquerel*.	Human, 3d day after delivery—*Simon*.	Cow—*Chevallier* and *Henry*.	Ass—*Chevallier* and *Henry*.	Ewe—*Chevallier* and *Henry*.
Water . . .	889·08	828·0	870·2	916·5	856·2
Sugar . . .	43·64	70·0	47·7	60·8	50·0
Casein and extractives	39·24	40·0	44·8	18·2	45·0
Butter . .	26·66	50·0	31·3	1·1	42·0
Salts . . .	1·38	3·1	6·0	3·4	6·8
TOTAL,	1000·00	991·1	1000·0	1000·0	1000·0

It will be seen from the above table that ewe's milk is the richest of all milks, while the milk of the ass is rich in sugar, but poor in butter and casein. It will also be observed that human and cow's milk are, on the whole, much alike, and hence the last may be substituted for the first. The human milk examined by Simon soon after delivery was rich in sugar and fat, the amount of the latter being nearly double that found in ordinary milk. Vernois and Becquerel found in the ash of human milk 6·9 per cent. of calcium carbonate, 70·6 per cent. of calcium phosphate, 9·8 per cent. of sodium chloride, 7·4 per cent. of sodium sulphate, and 5·3 per cent. of other salts.

Milk is often adulterated with water, flour, brain-substance, almond emulsion, chalk, &c. These adulterations may be readily detected by microscopical examination.

PARTHENOGENESIS.

By parthenogenesis (from παρθενία, virginity; γένεσις, generation) we understand the production of offspring, unlike their parents, which *may* take place without a true act of fecundation, that is, without at each birth, a necessary union of the male and female elements. This process had been previously called by Steenstrup, *alternate generation*. But many of the facts described under this term refer not so much to an alternate as to a continuous development. Thus, many insects spend part of their lives as a worm, and part as a moth. The moth produces the worm, and the worm produces the moth; but this is not an alternate, but a different phase of the same generation. So a

correct knowledge of the development of the *Medusa aurita* has shewn that what naturalists had considered to be four distinct animals are in fact only different stages in the development of one animal. The formation of the Aphis is especially alluded to by Steenstrup. Several of these insects are produced from the mother, and each may produce others, although it is only certain of them which become transformed into a fly. But the generation of a plant may be called alternate in the same sense as it is used in the case of the Medusa or the Aphis, inasmuch as the seed produces a root and a leaf-bud, which proceed to develop other leaves before they finally produce flowers with seed like that from which the plant originated. The term *parthenogenesis*, therefore, proposed by Owen, is the more correct one, and the following are examples of this process :

1. *The development of the Tape Worm, &c.*—The researches of Helminthologists, but especially of Siebold, Van Beneden, Dujardin, Steenstrup, and Blanchard, have cleared away the mystery which has so long hung over the origin of tape worms and other entozoa. It seems now determined that tape worms are only further stages of development of cysticerci, as flukes are only further stages in growth of certain cercariæ.

Professor Siebold first pointed out that the *cysticercus fasciolaris* found in the liver of the mouse, reaches its ultimate stage of development in the intestines of the cat, and is there transformed into the *tœnia crassicollis*. This fact was confirmed by a careful series of observations made by Dr Henry Nelson, who, in his thesis presented to the University of Edinburgh in 1850, carefully traces and figures all the various stages which the tape worm of the cat passes through. Each joint of this worm is estimated to contain 125,000 ova, which gives for the entire animal about 12,500,000. These minute bodies pass off by the fæces in incalculable numbers, and enter into the body of the mouse, mixed with its food or drink, or by licking its furry coat, to which they adhere. From the alimentary canal of the mouse they may enter the liver of that animal in three ways—1st. They may ascend the bile ducts. 2d. They may pass through the coats of the intestine, and penetrate the adjoining portion of the liver. 3d. They may bore their way into one of the mesenteric veins, and be carried by the blood along the vena porta to the liver. Dr Nelson considers the latter to be the most correct view, as he shews that the ova are furnished

with temporary teeth, which enable them to pierce the tissues. That they do not perforate the intestine, and so get into the liver, is shewn by the fact that they are most developed on the surface of that organ, and least so in its interior. Neither are they found especially in the biliary ducts, like the *distomata.* Hence the blood seems to be the channel of their introduction, an idea still further supported by facts, the number of which is rapidly augmenting, demonstrating entozoa in various stages of development existing in the blood itself. Arrived at the liver, these ova are transformed into *cysticerci fasciolares*, and would never proceed further in development in the mouse, but being eaten by the cat, they become tape worms, and are transformed into *tæniæ crassicolles.*

This series of observations renders it probable that all the various kinds of tœnia are only advanced stages of development of different cysticerci. Dr Nelson points out that "the head of the *cysticercus cellulosæ* resembles in every respect that of the *tænia solium* of man. The two figures given by Bremser are identical, if we allow for the stretching of the neck in the latter. Both have a double circle of hooks ; and although the *tœnia solium* is sometimes found without any teeth, Bremser has fully proved that this is the result of age, and not the original condition. He also observed, that as the worm increased in age, one row of the double corona first fell off, and was after a time followed by the other, leaving the worm thus unarmed." Besides, man feeds on animals in which these cysticerci are common, especially on the pig and sheep; and it has been observed that in countries where meat is often eaten raw, as in Abyssinnia, tape worms are very common. The reason of the rare occurrence of tœnia in civilised countries is probably owing to the cooking of food, which destroys the vitality of the cysticerci. Occasionally, however, it may easily be conceived that, owing to meat being very underdone, or to the tenacity of life in certain of these creatures (many of them resist a high temperature without injury), they may escape the action of the teeth, arrive living in the human stomach, and be converted into young tœnia.

These ideas with regard to the origin of tape worms have been converted into certainties by the experiments of Dr Küchenmeister.* He fed dogs and cats upon parts of animals which

* *Prague Vierteljahrschrift,* Band i. 1852, p. 126.

contained different kinds of cysticerci, and subsequently found the tape worms into which these had been transformed presenting various stages of development, according as the life of the animal which had eaten the cysticerci was more or less prolonged afterwards. Every precaution seems to have been used in these experiments, one of which may be cited. An old dog, during a period of from six to eight weeks, was frequently purged with castor oil, so as to prevent the possibility of tape worms being present. On the 18th of March 1851, he ate food containing ten cysticerci. On the 25th, he ate as many more, and on the 1st of April several others which were not numbered. On the 10th of April the dog was killed, and thirty-five tœnia were found in the intestines, of which five were from 124 to 390 millimetres (from about 5 to 15 inches) in length, and possessed from 130 to 160 joints. There were six others from 25 to 96 millimetres (1 to 5 inches) in length, having from 40 to 60 joints. There were twenty-one others which measured from 8 to 16 millimetres ($\frac{1}{4}$ to $\frac{1}{2}$ an inch) in length, in which the joints were so indistinct that they could not be counted. Lastly, there were three measuring from 4 to 5 millimetres (1-6th of an inch) in length, in which the joints could scarcely be distinguished. Considering the power of contraction and elongation possessed by these worms, their length was not so decided a character of their stage of development, as the size of the head and hooks, which corresponded to the three periods in which the cysticerci had been swallowed.

On feeding dogs upon the liver of the mouse, containing the *C. fasciolaris*, Dr Küchenmeister never found tœnia in the intestines. But when he fed cats on the same liver, the intestines contained the *tœnia crassicollis*. This observation indicates that not only are certain cysticerci transformed into certain tœnia, but that the former require certain habitats, or peculiar animals in order to undergo this transformation. Although the present amount of our knowledge does not enable us to state from what kinds of cysticerci many species of tœnia are formed, it seems tolerably certain from the observations of Siebold, Nelson, and Küchenmeister that the *Cysticercus fasciolaris* of the mouse is transformed into the *Tœnia crassicollis* of the cat; the *C. pisiformis* of hares and rabbits into the *T. crassiceps* of the fox; the *C. tenuicollis* of ruminantia and squirrels into the *T. serrata*, so common in the dog, and the *C. cellulosæ* of

the pig and sheep, into the *Tænia solium* of man. It is also tolerably certain from the observations of Eschricht that the *Bothriocephalus latus* of man found in certain countries, especially in Russia, is the further development of a species of *Ligula*, which exists in large numbers in the flesh of the dorse, and other fish of the northern seas. Dr Cobbold has shewn that the *T. medio-canellata* is, in fact, the most common tape worm of man, and that it infests the ox and enters the human family by the process of eating beef.* It has no circlets of hooklets, but four suckers.

The importance of the head of tape worms, so long recognised by practical physicians as the only certain proof of the complete expulsion of the worm, has also received an explanation from the researches of Helminthologists, into the anatomy and development of these animals. Notwithstanding the doubts expressed by Van Beneden as to the lateral canals being connected with the digestive system, and his notion of their being peculiar secreting organs, Dr Nelson in his thesis has distinctly traced them into the suckers of the *tænia crassicollis*. From each of the four suckers a canal descends, which afterwards unite, two and two, to form the lateral canals. He also carefully describes the manner of feeding and propulsion of the contents of these canals from the cephalic to the caudal segment. Hence the head is important as the means by which the animal is nourished.

But the head is also important, as pointed out by Van Beneden, as the part from which all the joints are thrown off, by gemmiferous reproduction, those formed first being pushed downwards, and being further developed as they go. Hence, why the joints are narrow near the head, and become larger and longer near the tail. The latter after a time separate, but according to Van Beneden, may still go on developing, and become, he thinks, a species of fluke or distoma. In fact, he considers a tape worm as a compound fluke worm, the whole consisting of three stages or periods: 1st, the cystic head (*Scolex*); 2d, the compound tape worm (*Strobila*); 3d, the separated joints (*Proglottis*). This latter view, however, is opposed by the observations of Steenstrup as to the development of the fluke, as much as by what we know of the arrangement of the nervous and digestive systems of this entozoon.

* Spencer Cobbold, Tapeworms. 1866. Appendix.

2. *The development of the animals forming coral reefs.*— These reefs are banks or walls of hard materials, growing from a definite depth to near the surface of the water. They are entirely the work of marine animals, belonging to the class of Polyps (sub-kingdom *Cœlenterata*). These form communities, growing up in the same way, budding side by side or dividing, and while so multiplying remaining united together, so as to form a larger and larger mass. The buds are so arranged that the ends have a hemispherical form, or appear like branches dividing and subdividing. This is owing to the manner in which the new individuals unite with one another. Each species of Polyp has its own peculiar mode of budding, branching, and ramifying, giving to it as distinct an appearance as exists among different trees. The number of these different species is very great, and they all have not only peculiar features and habits, but require different positions in the sea. There are those which are only found in shallow waters; others again in water two fathoms deep; others are never found in waters which are less than five or six fathoms deep; and others only in waters at least ten fathoms deep. These peculiarities are as marked among corals as the differences we observe in the distribution of plants and trees on mountain slopes, or among animals in different localities. The mere fact of the water being more or less clear is enough either to foster their growth or cause their destruction. The animal made of such soft and tender materials must be very nicely and evenly adjusted in its structure to be able to bear the pressure of a particular depth and no other. Hence why those who live near the surface cannot exist a few fathoms deep?

A coral reef, then, is a structure built up from a definite depth, successively and gradually, not by one kind of coral, but by a great variety of kinds, combining together and forming by their joint work a wall, which, from a given depth, may end in reaching the surface of the water. And while it is growing, this wall is all the time changing its builders. It is not one kind that commences and completes the structure to the summit. One kind does a part of the work, and then ceases; another kind comes in and continues the work for a while, and ceases in its turn; and so on, till it is completed.

Suppose we have a slanting shore, and that at 600 feet distance from the shore the depth is 10 or 12 fathoms, the animals

would commence building a wall, steep towards the ocean, slanting gently towards the shore, rising in the end towards the level of the water. The effect of muddy water, occasioned by storms and tides, raising the sand and mud at the shore, is to destroy the corals near the shore and prevent the building of the reef. On the other hand, where there is a somewhat steeply slanting shore, and the water is pure and plentiful, the conditions are most favourable to the animals. Consequently the side towards the sea will be built almost vertically, and will grow more rapidly than that towards the land.

It having been ascertained that different portions of the reef, at varying depths, are built by different species of corals, and that these are immovably attached together, the question arises, Whence did these new corals come which built the later portions? It is now known that this results from free polype ova becoming fixed and presenting simply various degrees in development as they attach themselves to different elevations on the reef. In each degree of development, however, a similar form may be reproduced without a new act of fecundation.

This story of the coral reef, as an example of parthenogenesis among animals, would not be complete without a reference to what it teaches us of the chronology of the earth. Now, Agassiz has determined that on the southern coast of Florida, within fourteen years, the addition in the way of a crust of corals found upon the foundations of Fort Taylor, does not exceed one inch in depth; therefore, less than a foot would grow in a century, and this he considers is overstating the rate of growth. It would take, therefore, six thousand years to produce a reef 60 feet high, which is the known depth of the inner coral reefs on that coast. But outside these are other reefs which must have grown since the internal ones were formed, because without the existence of the former, the coast could not have presented the conditions necessary for their production. But the outer reefs are as thick as the inner, and must also have taken six thousand years to grow. But an examination of the shore proves that it also is an ancient coral reef, and for the same reason must have been formed before the coral reefs nearest the coast; in that we have a third item of six thousand years to add to its chronology. Further, within the shore on the Indian hunting grounds are eminences called hummocks, which are also coral reefs concentric with the shore, and formed

of the same species of corals as those now building the internal reefs. So that, if there be any accuracy in these two leading facts, viz., that the rate of growth is less than a foot in a century, and that the existence of an outside reef precludes the formation of a reef inside, we have the evidence, in the existence of these four consecutive reefs, that twenty-four thousand years ago there was a sea washing the plain where those hummocks are, and that no reef had then formed beneath them.

Yet this is not all. These animals are of the same kind as those that live now, and what has been described occurs within a narrow track of fifteen or sixteen miles. Sixty miles in the interior is a lake, up to the shores of which the ground consists of similar hummocks formed of coral. Nay more, the whole peninsula of Florida is entirely made up of coral reefs, and if so, we can scarcely escape the assumption that hundreds of thousands of years must have been required for the animals to build it. And yet, according to Agassiz, this must be nothing compared to the age of the world, because it is a period within which the species of animals which now live existed on the earth ! That is to say, in the mind of a geologist, altogether modern time !*

3. *The development of bees.*—The observations of a distinguished Apiarian, M. Dzierzon, of Carlsmarkt in Silesia, confirmed by the scientific researches of Professor Siebold as to the reproduction of bees, offers another illustration of Parthenogenesis.

In the hive, as is well known, there are three sets of bees, the queen-bee, the drones or male bees, and the workers, which have generally been called neuters, but they are, in fact, imperfectly developed female bees. The queen or female bee is only impregnated once, by one of the drones, while in the air, during what is called the nuptial flight. On this occasion she receives into a receptacle the seminal fluid, which is sufficient for the remainder of her days, and her impregnation may be known at once (as the receptacle formerly contained limpid fluid, whereas afterwards it is white and opaque). The receptacle or cavity lies apart from the ovary, but communicates with the oviduct, from which, however, it can be shut off at will.

The workers having prepared three kinds of cells, viz., drone cells, worker cells, and royal cells, each of which has its own

* See Agassiz on "The Structure of Animal Life," 8vo, 1866, p. 52, *et seq.*

peculiar form, and size, the queen proceeds to deposit an egg in each, laying at the rate of perhaps 200 a-day, or 12,000 in two months. In doing this she takes care to bring every one of those destined for the royal and the worker cells into contact with the seminal fluid, but takes equal care to keep free from such contact every one of those destined for the drone cells. She is probably guided in this by the feel of the individual cells which she visits, the requisite proportions and number of which have been previously made by the workers. In due time the number of bees, increasing beyond the capacity of the hive to hold them, a swarm comes off, headed by the old queen, who leaves her place in the hive to be supplied by one of her royal daughters. The latter heads the next swarm, which commonly leaves the hive within from seven to nine days after the first, and during the nuptial flight, she is duly impregnated by one of the attendant drones. The like happens with the other swarms; and after the departure of the last, a struggle for the vacant throne ensues among the young princesses that remain in the hive, one of which succeeds in gaining it, while the others are all of them destroyed. The first thing done by the young queen is to get wedded, an affair seldom delayed beyond a day or two after her accession, and invariably celebrated in the open air, the queen leaving the hive for that purpose, accompanied by a crowd of drones, one of whom she selects, and returning to the hive after the consummation of the nuptials. Within a couple of days thereafter, she begins to lay, depositing the eggs with the precautions in respect of the seminal fluid already noticed, in order to maintain due proportion between the several sorts of bees.

But it occasionally happens unfortunately for the well-being of the community that from some defect in her wings incapacitating her for flight, a queen cannot leave the hive. Her nuptials, therefore, are not celebrated (and she remains a virgin). Possessing, however, all the fertility of a wedded queen, and the same propensity for reproducing the species, she proceeds to lay eggs notwithstanding, and none but drone bees resulting, the economy of the hive is subverted. Young workers and queens, it is true, make their appearance also; but they are the progeny of the old queen, who, just before her departure with the first swarm, had laid eggs which could be hatched only a little while in advance of those of her unhappy successor. As

soon, therefore, as one of these young queens becomes competent for duty, she attacks the reigning sovereign, and takes possession of the throne. Thereupon a general massacre of the redundant workers and drones takes place, and the body politic is again restored to its natural condition. Sometimes a hive has no queen at all. In that case Leuckart found that if a worker bee be fed with royal food it is transformed into a queen. This consists of a peculiar paste prepared in the digestive organs of the workers instead of pollen and honey.*

Such are the facts from which it follows that male animals in insects may be produced independently of the union of the ova with spermatozoids, in the same way that buds are thrown out in trees, or new polype heads are formed, without a distinct act of generation. Unimpregnated ova in insects are analogous to the successive buds in a tree, which are annually capable of being fertilised, although they are not all so. The queen, in this respect, is like a tree, uniting two modes of development—seminal or oviviparous, and gemmiparous. She is a female in form only, not in reality. She has the appurtenances of a female, but the attributes of a male ; and is, in fact, a male in a higher sense than even the drone. She is a *typical* bee, adequate of herself to reproduce her kind in its highest, which is the male form, and requiring only the co-operation of an ordinary male to reproduce herself in order to the production of those master bees which are the mainspring of the whole economy of the hive. Her ovary is, properly speaking, a *gemmarium*, the products of which she herself fertilises at will by shedding on the germs or ova the semen stored up in her receptacle.

4. *The development of the aphides, or plant lice.*—In 1745, Bonnet directed the attention of physiologists to the development of the aphides or plant lice, a process of true parthenogenesis.† At the close of summer the impregnated ova of the aphis are deposited in the axils of the leaves of the plant infested by the insect, and these ova are hatched the following spring, a wingless six-footed larva being produced. These larvæ will produce a brood of eight wingless larvæ like themselves without any connection with the male. No winged males are to be

* See Siebold, On Parthenogenesis, translated by Dallas, 1857 ; also Harvey, On the Seed and the Bud, pp. 45, 6, 7.

† Bonnet: "Traité d'Insectologie, ou Observations sur les Pucerons," 1745.

found at this period. This procreation from a virgin mother will go on to the eleventh generation before the influence of the original impregnation has been exhausted. At this period, however, individual growth obtains the mastery over the reproductive capacity, and some members of the last brood are changed into winged males, while others become ordinary egg-bearing females. These latter are impregnated by the males, and then the process is repeated the following year.*

Natural Selection.

But while that peculiarity of reproduction, named parthenogenesis, is of great importance in enabling us to understand the numerous varieties of forms which exist among the invertebrata, another principle—that of natural selection—has recently been referred to by Mr Darwin,† which helps us to account for the origin of species and the variations which exist among animals, as the result of sexual generation. We have previously alluded to the fact that the external characteristic form is generally determined by the male, while the bulk and internal qualities are dependent on the female. Of this there are many examples.

1. *Hybrids between the horse and the ass.*—A male ass and a mare produce a *mule*, while a stallion and a female ass produce a *hinny*. In the first case, the ears are long, the tail tufted, the feet small, and the animal brays. In the second case, the head is that of the horse, with short ears, the legs coarse and strong, and the animal neighs.

2. *The case of the otter sheep.*—Another remarkable example is that of the Ancon, or otter sheep. "A farmer in Massachusetts possessed a flock of fifteen ewes and a ram of the ordinary kind. In the year 1791, one of the ewes presented her owner with a male lamb, differing from its parents by a proportionally long body and short bandy legs, whence it was unable to emulate its relations in those sportive leaps over the neighbouring fences, in which they were in the habit of indulging, much to the good farmer's vexation. His neighbours imagined that it would be an excellent thing if all his sheep were endued with the stay-at-home tendencies inforced by nature upon the newly-

* Owen, On Parthenogenesis, p. 24. 1849.

† Darwin, On the Origin of Species by means of Natural Selection, or the Preservation of Favoured Races in the Struggle for Life.

arrived ram, and they advised Wright to kill the old patriarch of his fold, and instal the Ancon ram in his place. The result justified their sagacious anticipations. The young lambs were almost always pure Ancons, or pure ordinary sheep ; and when sufficient Ancon sheep were obtained to interbreed with one another, it was found that the offspring was always pure Ancon." In this well-authenticated instance we have a distinct race established at once, or by a leap, and that race breeding true. When the Ancon sheep were herded with other sheep they kept together, so that it was believed that this breed might have been indefinitely protracted, had it not been superseded by the introduction of the Merino sheep, which were not only superior to the Ancons in wool and meat, but were equally quiet and orderly.*

3. *The case of Gratio Kelleia.*—A Maltese of the name of Gratio Kelleia was born with six fingers upon each hand, and six toes upon each foot. He married a lady having the usual number of fingers and toes. The result of the marriage was four children : the first, Salvator, had six fingers and six toes ; the second, George, had five fingers and toes, but one was deformed ; the third, Andrè, had five fingers and toes, perfect ; and the fourth, a girl christened Marie, had five fingers and five toes, but her thumbs were deformed. These all grew up and married five-fingered and five-toed individuals. " Salvator had four children ; they were two boys, a girl, and another boy : the first two boys and the girl were six-fingered and six-toed like their grandfather ; the fourth boy had only five fingers and five toes. George had only four children : there were two girls with six fingers and six toes ; there was one girl with six fingers and five toes on the right side, and five fingers and five toes on the left side, so that she was half and half. The last, a boy, had five fingers and five toes. The third, Andrè, you will recollect, was perfectly well formed, and he had many children whose hands and feet were all regularly developed. Marie, the last, who, of course, married a man who had only five fingers, had four children : the first, a boy, was born with six toes, but the other three were normal. . . . . Now, what would have happened if these abnormal types had intermarried with each other—that is to say, suppose the two boys of Salvator had taken it into their heads to marry

* Huxley : Westminster Review, 1860. Article on Darwin, " On the Origin of Species."

their first cousins, the two first girls of George, their uncle? You will remember, that these are all of the abnormal type of their grandfather. The result would probably have been, that their offspring would have been in every case a further development of that abnormal type. You see it is only in the fourth, in the person of Marie, that the tendency, when it appears but slightly in the second generation, is washed out in the third; while the progeny of Andrè, who escaped in the first instance, escape altogether." *

4. *The case of Lambert, the porcupine man.*—Another example of hereditary transmission is the remarkable case of Edward Lambert, "the porcupine man." † In 1755, he was about forty years of age. His skin seemed like a "dusky-coloured thick case, exactly fitting every part of his body, made of a rugged bark or hide, with bristles in some places." . . . . "The bristly parts, which were chiefly about the belly and flanks, looked and rustled like the bristles or quills of a hedgehog shorn within an inch of the skin." The remarkable point in this man's story, reports Mr Henry Baker, is, "that he had six children, all with the same rugged covering as himself; the first appearance whereof in them, as well as in him, came on in about nine weeks after the birth. . . . . It appears therefore past all doubt, that a race of people may be propagated by this man, having such rugged coats or coverings as himself; and if this should ever happen, and the accidental original be forgotten, 'tis not improbable they might be deemed a different species of mankind: a consideration which would lead one to imagine, that if mankind were all produced from one and the same stock, the black skins of the negroes, and many other differences of the like kind, might possibly have been originally owing to some such accidental cause." This man evidently suffered from a disease termed Icthyosis (ἰχθύς, ἰχθυος, a fish), an affection of the skin which he transmitted to his children, and according to another authority, to two grandchildren.

The principle of natural selection has been extensively applied by Mr Darwin to explain the origin of species. According to him, every animal and plant is capable of increas-

* Huxley "On our Knowledge of the Causes of the Phenomena of Organic Nature. Six Lectures to Working Men." 1863. Pp. 95-97.

† "Philosophical Transactions," vol. xiv. No. 160, 1730, and afterwards in vol. xlix., part i., for 1755.

ing with such rapidity, that if it were unchecked by other species, it would soon occupy the greater part of the habitable globe. But in the *struggle for existence*, few only of those which are brought into the world can obtain food and arrive at maturity. It must be evident that the weak succumb and the strong remain. We have the *survival of the fittest.* Now, slight accidental causes may occasion variations, which being transmitted become more and more marked and permanent, so that the various conditions which influence life—such as climate, locality, food, &c.—may influence the species of animals. In any given species, those alone survive which have some advantage over the others, and this is often determined by a slight peculiarity capable, in a severe competition, of turning the scale in their favour, such as the possibility of procuring food during the least favourable seasons, and of escaping the attacks of their most dangerous enemies. These peculiarities are transmitted, and thus we have a species. We have many admirable examples of the principle of selection in breeding, such as—

1. *The varieties of dogs.*—The numerous varieties of dogs, from the Newfoundland or St Bernard to the lap-dog or terrier, are believed by Darwin to be produced from not more than two or three wild species. John Hunter maintained that the wolf, the dog, and the jackal were all of one species, because he had found, in two experiments, that the dog would breed both with the wolf and the jackal; and that the mule, in each case, would breed again with the dog.

2. *The varieties of pigeons.*—From the common rock pigeon (*Columba Livia*) no fewer than 150 distinct races of domestic pigeons have been produced. These all have received names and breed true; and at least twenty of them would be classed by an ornithologist, if he shot them in the woods, as well-defined species; while extreme forms, such as the short-faced tumbler, the pouter, and the fan-tail, would not be placed in even the same genus as the rock pigeon. Among pigeons, pigeon fanciers can produce, by cross breeding, not only differences in outward form, but they find even the number and size of many of the bones changed.

Atavism.—It is remarkable that now and again we find in the offspring of all tame pigeons some peculiar characteristic of the rock pigeon, especially in the plumage. This reappearance

of marks of an old progenitor is termed *atavism* (*atavus*, an old grandsire). Similar facts have been observed among other animals.

Sexual Selection.

Other circumstances have recently been pointed out by Mr Darwin as producing permanent differences in form among men and animals.* He believes that the sexual instincts and passions have a powerful influence. For example :

1. *Law of battle.*—Among animals there is a *law of battle*, that is, the males fight for possession of the female, and any male possessing a peculiarity giving him an advantage in the contest over his opponent, obtains the female, and in accordance with the law already stated (p. 415), transmits this peculiarity to his descendants. This holds good to the present day among barbarous nations. "There can be little doubt," writes Mr Darwin, "that the greater size and strength of man, in comparison with woman, together with his broader shoulders, more developed muscles, rugged outline of body, his greater courage and pugnacity, are all due in chief part to inheritance from some early male progenitor. These characters will, however, have been preserved or even augmented during the long ages whilst man was still in a barbarous condition by the strongest and boldest men having succeeded best in the general struggle for life, as well as in securing wives, and thus having left a larger number of offspring."†

2. *Voice.*—According to Mr Darwin, voice and musical powers are influential in reference to the propagation of the species. Among all Mammals, the males use their voice more during the breeding season than at any other time. The females use their voice as a love-call. The male Gibbon has a loud musical voice, and according to Owen, this anthropomorphic ape "may be said to sing." Mr Waterhouse thinks it highly probable it utters its musical notes during the season of courtship. "Women are generally thought to possess sweeter voices than men, and as far as this serves as any guide, we may infer that they first acquired musical powers in order to attract the other sex. But if so, this must have occurred long ago, before the progenitors of man had become sufficiently human to treat and value their women merely as useful slaves. The impassioned

* Darwin, "The Descent of Man and Selection in relation to Sex," 1871.

† "Descent of Man," &c., p. 325.

orator, bard, or musician, when, with his varied tones and cadences, he excites the strongest emotions in his hearers, little suspects that he uses the same means by which, at an extremely remote period, his half-human ancestors aroused each other's ardent passions during their mutual courtship and rivalry." *

3. *Idea of the beautiful.*—It is well known that different races of mankind have different ideas of personal beauty, and that great care is taken of those individuals possessing it. Thus peculiarities are intensified and transmitted through many generations till they become permanent. Sexual selection has effected much in producing the high style of beauty found among the aristocracy of civilised nations. Members of wealthy families, in which primogeniture has long prevailed, chose from all classes the most beautiful women as their wives, and consequently their descendants become more handsome. Among certain tribes of negroes, according to Winwood Reade, the ugly women are continually eliminated and sold as slaves, a practice which, though morally wrong, has produced tribes remarkable for their "uniformly fine appearance."

Among men, both savage and civilised, many causes interfere with the action of sexual selection, but we know enough to confirm the opinion that in this matter man obeys the same laws as regulate inferior animals.

Such is a very brief account of certain of Mr Darwin's ingenious opinions. No one can peruse his works without recognising their high scientific value and the large number of facts brought to bear on the inquiry. Very few of those who ridicule Mr Darwin's ideas have ever read his books and examined the evidence he furnishes in support of his doctrines.

It has long been known that many diseases are hereditary; but it has recently been pointed out by Brown-Séquard that epilepsy induced in Guinea-pigs is often transmitted to their offspring, even when produced artificially. He has found that in these animals a morbid condition of the nervous system is caused by section of one lateral half of the spinal cord, or of the posterior columns reaching to the cornuæ of grey matter, or of the sciatic nerve, and also in some cases by the simple puncture of the spinal cord. Under any of these conditions, true epileptic attacks may be readily produced by pinching the skin of the cheek. Many Guinea-pigs survive the operation and breed; and in the young, convulsive attacks may be produced

* "Descent of Man," p. 337.

in the same way. Here we have a disease artificially produced, yet, nevertheless, faithfully transmitted.*

HETEROGENESIS.

By heterogenesis (from ἕτερος, diverse, different ; γένεσις) we mean the production of living beings without parents.†

History.—This subject has engaged the attention of physiologists since the days of Aristotle. He, as well as most of the naturalists who lived previous to the time of Harvey, were of opinion that dust, decomposed flesh, and other dead substances might, under the influence of heat, air, and water, give rise to vital organisms. This mysterious process, by a singular perversion of language, has been called spontaneous generation. Francis Redi, a physician of Florence, was the first who clearly demonstrated, in 1638, that the larvæ and worms found in a dead body were not produced by putrefaction, but originated from flies' eggs deposited in the flesh.‡ The researches into generation, of William Harvey, led him to announce the law, "Omne vivum ex ovo." Since his day the belief has been general, that all animals and plants are derived from eggs or seeds; that vitality is always transmitted, and never created; and that, where these fundamental principles cannot be recognised, the minuteness of the germs and their wide diffusion throughout nature, and more especially in the atmosphere, offer a sufficient explanation of what may appear mysterious. Nature, it was argued, must be uniform in her operations, and analogy warrants our supposing that the same law of generation which applies to the higher animals and plants, is equally applicable to the lower.§

* Brown Séquard: Archiv. gén. de médecine. Fevrier, 1856. Vol. vii. p. 143. Mars-Avril, 1869, p. 211.

† Dr Bastian proposes the word Archebiosis (ἀρχή, beginning, and βιός, life) to express the production of living organisms from non-living materials, while Heterogenesis, according to him, is the production of living beings from pre-existing organisms living or dead. But all living creatures originate from organic matter which has once lived, such as the material dissolved in an infusion, according to the law of molecular organisation as explained (p. 46). I therefore regard it as better to define the terms Homogenesis, Parthenogenesis, and Heterogenesis, as they will be found in the text.

‡ "Experimenta circa Generationem Insectorum."

§ Most of the arguments on the other side, that is, in favour of Heterogenesis, will be found well stated by Burdach ("Physiologie," tome i. p. 8, *et seq.*)—arguments admitted by Allen Thomson "to throw the balance of evidence in favour of the spontaneous production of Infusoria, mould, and the like." (Todd's "Cyclopædia," article—Generation, vol. ii. p. 430. 1839.)

The ova, seeds, and primary cells, however, which were supposed to be floating in the atmosphere, though constantly looked for, even with the most powerful modern microscopes, can nowhere be found, and more careful investigation of the numerous forms of life which spring up in putrescent and fermented fluids have utterly failed in connecting them with pre-existing germs. Many scientific men, therefore, who had personally investigated the subject, were once more led into the belief in an equivocal or doubtful generation of the lowest forms of animal and vegetable life. This belief was strengthened by the appearance, in 1845, of a Memoir * by M. Pineau, carefully describing the evolution of organisms in the pellicle on the surface of infusions, and more especially in 1859, of a remarkable work by M. Pouchet, entitled "Heterogenesis, or Spontaneous Generation," as distinguished from Homogenesis, or Generation from Parents. This book contains numerous original experiments and observations made by the author, proving, as he thinks, that infusoria originate in a finely molecular, or, as he calls it, proligerous pellicle on the surface of decomposing fluids, without pre-existing cells or germs of any kind, and therefore independently of parents.

The publication of this book has led to a controversy which has continued up to the present moment. The theory of atmospheric germs, or that of the Panspermatists, has been sustained by M. Pasteur, who, by new experiments, has revived the doctrine that fermentation and putrefaction are not chemical processes, as has been maintained by Liebig, but physiological phenomena dependent on living germs derived from the atmosphere. These experiments have all, however, been since repeated by Pouchet and others, and their accuracy, as well as the correctness of his conclusions, are by them utterly denied. An extraordinary amount of research, ingenuity, and talent has been displayed by the advocates on either side, the results of which will be found recorded in numerous communications printed in the "Comptes Rendus" of the proceedings of the Academy.

Before stating more particularly the arguments advanced by the controversialists, on one side or the other, I propose describing shortly the results of some investigations undertaken by

* "Recherches sur le developpement des Animalcules," etc. Ann. des Sc. Nat. (Zoologie) tom. iii. p. 182, and tom. iv. p. 103.

myself, with a view of determining precisely the facts of the case, and the nature of the phenomena which have excited so much discussion. In October 1863 I commenced a series of observations, with the aid of my former assistant, Dr Argyll Robertson, which were directed (1st) to determine with exactitude, by means of the microscope, the changes which occurred on the surface of infusions during the development of plants and animals there; and (2d) to ascertain what influence the air treated in various ways exercised on such growths. These observations were carefully repeated and extended in October 1864. They were repeated, and numerous other experiments performed, with the aid of my late assistant, Dr Rutherford, in 1868; and my present assistant, Dr M'Kendrick, has also investigated the subject, and varied the experiments. These investigations naturally divide themselves into (1st) observations by means of the microscope as to the development of infusoria; and (2d) experiments directed to destroy the supposed germs in the atmosphere, so as to prevent putrefaction.

1. *Mode of development of infusoria.*—This has already been described in detail (see p. 46).* Suffice it to say, in any vegetable or animal infusion there is first formed on the surface the *proligerous pellicle of Pouchet*, consisting of minute molecules. Two or more melt together to form a *bacterium*, and several bacteria unite end to end to form a *vibrio*.

The movements visible in the molecules and filaments vary according to the amount of development. At first the molecules which float loose in the fluid exhibit gyrations which cannot be distinguished from Brunonian movements. When short bacteria are formed, these exhibit peculiar vibrations,—often turn round on their own axis in various directions, and slowly change their place. They rarely dart rapidly through the fluid, or exhibit a serpentine motion. But when the vibrio is formed, the filament is pushed forward with greater or less velocity, at first presenting a wriggling, but, as it becomes longer, a more decided serpentine motion. A distinct flexure

* The powers I have employed for the investigation were an excellent lens of one-eighth of an inch focus, by Ross; but I also frequently used a one-twelfth of an inch by the same maker, and the immersion lens No. 10 of Hartnach, with varying powers of from 600 to 800 diameters linear. Occasionally I confirmed my observations with a lens made for me by Messrs Powell & Lealand, of one-twenty-fifth of an inch focus, whereby I obtained an enlargement varying from 1250 to 2000 diameters linear.

can be seen at certain points in the filaments, between the groups of molecular chains or filaments. Dumas says he has seen the molecules and bacteria uniting endways, a statement the correctness of which Pouchet doubts.* On two occasions, however, I was so fortunate as to see this occurrence.

Pouchet thought that the vibriones exuded a mucous matter, whereby one stuck to the other. If so, such exudation can only be poured out at their extremities, as they only unite lengthways, never crossways. I feel satisfied, however, that the reason the actual union has so seldom been seen is, 1*st*, That it only occurs at certain periods of development, and can only be followed by the eye when the movements are slow; 2*d*, That amidst such a multitude of minute moving bodies it requires a long time before two can be found exactly on one plane, and can be brought so accurately into focus that they can be watched for a sufficient time. Having, however, in two instances, actually seen the coalescence, I can have no doubt whatever that such is the true method of elongation. Numerous other facts seen among elongated vibriones support this view.

The further development which takes place from histogenetic and histolytic changes in the proligerous pellicle, resulting in the production of various forms of animal and vegetable life, has been previously described (see p. 47). These are dependent on temperature, season of the year, exposure to sunlight, and nature of the infusion. In all these cases no kind of animalcule or fungus is ever seen to originate from pre-existing cells or larger bodies, but always from molecules.

According to Dr Bastian, in the proligerous pellicle composed of bacteria, embryonal areas gradually appear. As a result of segmentations in these, specimens of *Monas lens*, 1-3300th in diameter, more or less suddenly make their appearance; they increase in size, occasionally assume an amœboid appearance for a time, and are ultimately transformed into real amœbæ. A membrane is formed around them and they become encysted, and in the interior of some of them there springs up a progeny of new bacteria, the production of which occasions their final dissolution.† He also describes the formation of numerous fungi occurring in the pellicle at the same time.‡ That we

* Nouvelles Expériences, p. 115.

† Proceed. Royal Society, March 21, 1872. Vol. xx., p. 250.

‡ Bastian, The Beginnings of Life. Vol. ii. p. 232. 1872.

should sometimes have animalcules, and at others fungi, is a well-known fact, the exact causes or conditions producing which are not yet explained. The Panspermatists, of course, are of opinion that the germs in the atmosphere are of many kinds, and that as they fall into various infusions they produce different results, in the same manner that varieties in ova or seeds develop themselves in peculiar localities or special soils. This assumption, however, seems to me opposed by the following experiment :—

If an infusion be placed in a deep glass vessel, which again stands in the centre of a shallow vessel containing the same infusion, and the whole covered with a large bell glass, it will be found in eight days that on the surface of the former are numerous ciliated animalcules, while on that of the latter only bacteria and vibrios exist. The experiment may be reversed, for if the shallow vessel be filled to the brim, and the deep vessel has only its bottom covered, then the ciliated microzoa will appear in the former, and the non-ciliated in the latter.*

As a result of these experiments, Pouchet has formularised a law to the effect that the production of ciliated animalcules is in an inverse ratio to the square of the surface, and that the production of monads is in a direct ratio to the cube of the mass of the same fluid.† To this law I have met with some exceptions, animalcules having been produced in some of our recent experiments in the shallow dish, and vegetations in the deep vessel, and *vice versa*.

It is difficult to explain how germs falling from the air on the same infusion, under identically similar conditions, with the exception that the fluid is in vessels of different forms, can vary the results. Whereas the fact that the higher infusoria are formed secondarily out of the disintegrated mass of the simpler ones, which can only take place where that mass is considerable, and floating on the surface of deep fluids, directly confirms the molecular theory of growth, and offers an illustration of how successive disintegrations give origin to different formations ‡ (p. 46).

That the infusoria originate and are developed in the molecular pellicle which floats on the surface of putrefying or fermenting

* Pouchet's Nouvelles Expériences, &c., pp. 135, 243-245. Paris, 1864.

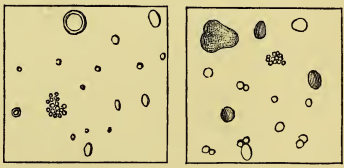
† Nouvelles Expériences, p. 134.

‡ See On the Molecular Theory of Organisation, by the Author. *Proceedings of Royal Society of Edinburgh*, 1861.

liquids, has been admitted by all who have carefully watched that pellicle with the microscope, more especially by Kutzing,* Pineau,† Nicolet,‡ Pouchet,§ Jolly and Musset,|| Schaafhausen,¶ Mantegazza,** and Bastian.†† The question therefore is, Are the molecules that constitute that pellicle derived from the air or the fluid,—are they precipitated from above, or do they float to the surface from below, like the globules of the milk which produce cream?

Now, it was in consequence of having professed to demonstrate what had escaped all previous observers—viz., the germs in the air—that M. Pasteur has made his name so famous. He tells us ‡‡ that he did this by causing a current of air to pass through a glass tube in which a pledget of gun-cotton had been placed. This was then dissolved in ether, and the sediment allowed to collect in a watch-glass. This sediment, after being repeatedly washed, and allowed to remain in distilled water for twenty-four hours at a time, is allowed to dry. A portion of the dried matter is then put upon a slide moistened with a weak solution of potash, and, being covered with another glass, is examined with the microscope. The results he has figured; and, very properly, he has given the scale of magnifying power under which they were drawn (Fig. 5), and which, by careful measurement, I have ascertained to be 180 times linear. His drawings, carefully copied, are represented (Figs. 1 to 4).

Fig. 1. Fig. 2.



* See Schaaffhausen, Comptes Rendus, tom. liv. p. 1046.

† Annales des Sciences Naturelles, 3me série, tom. iii. p. 182. This observer thinks he saw disintegrated fibres of meat and of other substances formed directly into vibriones,—in this he was incorrect.

‡ Arcana Naturæ, tom. i. p. 2.

§ Hétérogénie, p. 353. Nouvelles Expériences, p. 111.

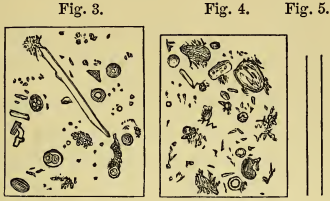
|| Comptes Rendus, tom. i. p. 934. ¶ Ibid., tom. liv. p. 1046.

** Institut Lombard, 1852, tom. iii.

†† Beginnings of Life. London. 1872.

‡‡ Annales des Sciences Naturelles, 4me serie, tom. xvi. p. 25.

Fig. 3. Fig. 4. Fig. 5.



Exact copies of the figures given by M. Pasteur of the dust he collected on gun-cotton, magnified 180 diameters. These should be compared with Fig. 10 (Plate II.), magnified 800 diameters, shewing what is seen to take place when infusoria are forming. Fig. 5, scale of one hundreth of a millimètre.

He says Figs. 1 and 2 represent organised corpuscles from dust collected in twenty-four hours, from the 16th to 17th November 1859. The manner in which these drawings, giving the volume and outline of the bodies, were made, is as follows: "After the dust has been prepared in the manner described, I took a portion of it from the watch-glass, and diluted it with a solution of potash, consisting of 5 parts of potash in 100 of water. As soon as I perceived a globule evidently organised under the microscope, I drew it. This is how figure 4 was drawn."* This description leaves it uncertain whether an exact copy was taken of any portion of the field of the microscope, and, therefore, whether the figure represents the exact number of corpuscles present, and their relation to each other. It only gives their form. But, assuming that the same kind of demonstration was made in each case, we have the relative numbers of these bodies taken from the gun-cotton in Fig. 1. Fig. 2 is another demonstration of the same after the addition of an aqueous solution of iodine. Fig. 3 represents the organised corpuscles associated with amorphous particles obtained on the 25th and 26th of June 1860. Fig. 4, the dust of an intense fog in the month of February 1861. In all these demonstrations he admits the organised corpuscles are comparatively scarce, because, he observes (p. 31), it is frequently necessary to change the field in order to see one of them, whilst at other times several could be seen together.

M. Pasteur thinks that these drawings indicate the number of organised corpuscles that may be arrested in a small mass of

* Annales des Sciences Naturelles, 4me série, tom. xvi. p. 25.

cotton through which 1500 litres of air, in one of the less-frequented streets of Paris, have passed in twenty-four hours, about three or four yards from the ground. These he estimates at several millions in a litre (p. 29).

Now, it must be remembered that M. Pasteur is a chemist, and it will be admitted by every histologist that no method could be more unsatisfactory for determining either the nature or the number of the corpuscles than the one he adopted. The solution of the cotton in ether, the frequent soakings in water, the desiccation, and then the addition of a solution of potash, must completely alter the character of any living corpuscles in the atmosphere. Then the forms he assumes to be organic are not necessarily so. They are exceedingly frequent among mineral substances, and siliceous rounded forms are common, which, of course, resist sulphuric acid.

Nature of dust.—Numerous investigations have been made, both before and since M. Pasteur wrote, to determine the nature of dust floating in the atmosphere—of that dust, for example, which a ray of sunlight reveals to us, when admitted into a chamber. It consists, for the most part, of different kinds of starch corpuscles; the debris of clothing, especially filaments of cotton, silk, and wool; the results of different kinds of combustion, whether of coal or of wood; various mineral bodies, globular or ovoid, amorphous or crystalline; and minute fragments of insects and vegetables; very rarely small seeds and microscopic animalcules.

These constituents vary to such an extent in different localities, as to enable the observer, in some cases, to determine whence the dust was collected. Starch corpuscles abound in the neighbourhood of flour-mills and bakeries; fragments of clothing where there have been crowded assemblies of persons, cotton and wool being predominant if the persons belong to the poorer classes, and silk if the upper classes have been present; the products of combustion predominate in smoky localities; mineral particles on the roads and highways; seeds, fragments of vegetables and insects, in market places, gardens, &c. &c. But although these constituents of the air vary in different places, infusoria, produced in all of them, are identically the same.

This has been tested in various ways. The dust has been

* Pouchet's Nouvelles Expériences, p. 73, *et seq.*

ransacked to discover organic germs,—collected and carefully examined with the microscope, near the soil, and on the summits of the highest buildings, not only in frequented, but in desert places; in crowded assemblies, as well as in empty Gothic cathedrals and ancient vaults—in the ancient palace of Karnack, on the banks of the Nile, in the tomb of Rhamses II. at the extremity of the Desert, as well as in the central chambers of the great pyramid of Ghizeh. The chief element of the dust collected in these places has been found to be starch corpuscles.* Large quantities of air have been drawn through tubes by aspirators, and collected on cotton, in distilled water, or projected on glass. The feathery snow, which, falling through the atmosphere, may be well supposed to collect its contents, has been melted, and the precipitate carefully collected. The emanations of marshy places, such as those of the Maremma in Tuscany, have been specially investigated.† The larynges and mucuous pulmonary surfaces of numerous animals have been explored, even to the inmost bone cavities of birds. On the summit of Mont Blanc, amidst eternal snow; on the glaciers of the Jura and of the Pyrenees, and in the deep crevasse ;‡ on the burning plains of Egypt, and in the markets of Constantinople, the dust of the atmosphere has been microscopically examined,—and in all with a like negative result as to the existence of germs. Nowhere could they be seen, or if a few, in the opinion of some, were visible, could they in any way account for the multitude of minute infusoria, which, in all these localities, not only readily spring up in putrid fluids, but in every instance are identically the same.§

Histological proof that infusoria originate in the coalescence of molecules formed in fluids.—From what has been previously said it follows that the pellicle that may be seen to form on the surface of infusions cannot possibly be derived from the dust of the air, as it bears no relation whatever in structure to dust. Nor can it be derived from the bodies figured by M. Pasteur as existing in the atmosphere. (See Figs. 1, 2, 3, 4, p. 426.) For how, it may be asked, could these bodies produce the incalculable

* Pouchet's Hétérogénie, p. 446.

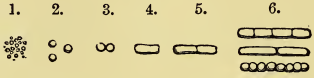
† L. Gigot's Récherches expérimentales sur la Nature des Emanations marécageuses, Paris, 1859. Récherches sur l'Air des Maremmes de la Toscane, par M. E. Bechi. Comptes Rendus, tom. lii. p. 825.

‡ Comptes Rendus, tom. lvii. p. 558.

§ Nouvelles Expériences, p. 75.

millions of minute molecules in the smallest fragment of the proligerous pellicle we can transfer to our microscopes, in which, as we have seen, the infusoria originate (p. 46)? It has been supposed that on falling from the air, they undergo rapid division, and spread over the surface with the greatest rapidity; but no one has ever seen this remarkable phenomenon, and the slightest consideration must shew that such an assumption is completely

Fig. 6.



Stages in the development of vibriones—800 *diameters linear.*

adverse to what can be readily demonstrated on the surface of every infusion. This histological argument merits special attention, because I do not see how it can possibly be answered. There can be no doubt that the minute molecules are formed first, and the bacteria, vibrios, and filaments, last. Supposing that the primary molecules, figured No. 1 (Fig. 6), enlarge to a certain point, No. 2, and then divide, how is it possible to explain the formation of elongated filaments at all? Surely the idea of their rapid multiplication by division is opposed to that of their power of elongating into bacteria and vibrios, whether by aggregation or growth from their extremities. It may frequently be seen that No. 3 is composed of molecules of exactly the same size as No. 2, which are floating loose,—a fact in favour of their coalescence rather than of their division, as then they would be reduced to half the size. It is more probable that although the smaller molecules may increase by imbibition of fluids, they have yet a constant tendency to aggregate together and melt into one another. No. 3 is not a proof of No. 2 dividing, but of two molecules coalescing; and when they unite, they form No. 4. Two or more of these uniting, form Nos. 5 and 6. When a similar process to this goes on in mineral bodies, as shewn by Mr Rainey,* it cannot suggest division but union (p. 41, Plate II. fig. 3; and this for the obvious reason, that the former would lead to disintegration, whereas, it can be seen in one case as in the other, that development is the result. In short, in the same manner as a tube is formed by a coalescence of cells, so is this

* On the Mode of Formation of Shells, &c., 8vo, London, 1855, p. 12.

minute vibratile vibrio formed by the coalescence of molecules. It may be argued, however, that each molecule elongates itself —that is, No. 2 is converted into No. 4; this into Nos. 5 and 6; and that No. 3 are sporules or ova, caused by the disintegration of No. 6. But this view is opposed by the fact that Nos. 1, 2, and 3 are seen *before* Nos. 4, 5, and 6 are produced. Of this all have satisfied themselves who have examined animal and vegetable infusions; and the conclusion, therefore, cannot be resisted, that the vibrios are derived from the molecules, and not the molecules from the vibrios.

But it may also be supposed, that while some have the property of dividing, others are capable of elongating or aggregating; this view, however, is not only opposed to observation, but is at variance with all that we know of embryonic development in plants and animals. When a plant consists of a single structural element, such as a cell or a tube, it will, I think, be admitted that growth in the sense of increased bulk, and growth in the sense of multiplication of its parts by division, do not proceed at the same moment of time. Every plant and animal follows, in this respect, the same law. Nutrition is carried on up to a certain point of maturity, and then, and not till then, does generation, or the separation of parts to form new creatures, take place. When plants and animals are complex in their structure, one organ or segment may be growing, while another is disintegrating; but in elementary parts there is a period for growth and reparation, and a period for division or separation. Hence, it seems to me, I am correct in thinking that if the primary molecules on the surface of an infusion possess the property of dividing, they cannot also, at the same moment, possess the property of elongating and forming filaments. The one function is subversive of the other. While, then, a cell or a vibrio may possess the property of growth and division, these two functions must be exercised at different periods of time,—so that, in reference to the early stage of formation, if the molecules divide, bacteria, vibrios, and filaments could not be formed. A mass of vibrionic molecules is not a compound organism; it is a mere aggregation of similar simple elements. Each of these in passing through certain phases of development may be arrested, or reach maturity at various periods, so that we frequently see different forms present at one time; but that the same elementary forms and the same stages of growth should

exhibit directly opposite functions, is surely not in accordance with physiological knowledge.

The conclusion we must arrive at therefore is, that the molecules seen on the surface of infusions out of which animalcules and fungi are produced, are not derived from the air.

Neither can they be supposed to pre-exist in the fluid, as then they would be readily seen, which they never are at the commencement. On this point nothing can be clearer than the microscopical evidence, so that it results from the facts and arguments which have been stated, that the more simple infusoria do not originate from cells or minute germs at all, whether in the atmosphere or in the fluid. This is the almost universal conviction of histologists who have carefully investigated the matter.

2. *Chemical Experiments which have been directed to destroy the supposed germs in the atmosphere, so as to prevent fermentation and putrefaction.*—Schutze, in 1837,* after heating an infusion to the boiling point, connected it with two of Liebig's bulbs, one containing sulphuric acid, and the other concentrated solution of potash. The air forced through these liquids he thought capable of destroying the atmospheric germs.

Schwann also, in 1837,† forced air, with the same view, through metallic tubes heated to redness; and found, when so calcined, it occasionally prevented infusorial growth. He thought that oxygen alone was not the cause of fermentation, but some substance in the air capable of being destroyed by heat.

Schræder and Dusch, in 1859, filtered the air through cotton before bringing it in contact with organic fluids. They found that some did, and others did not, undergo putrefaction, and were induced to believe that the presence of oxygen, and the formation of an acid, were the cause of fermentation. Schræder afterwards found that the yolk of an egg, milk, and the juice of meat without water, putrefied in air filtered through cotton, and supposed it to contain an active substance, the nature of which was unknown. ‡

* Poggendorf's Annalen, 1837, p. 41; and Edinburgh New Philosophical Journal, 1837.

† Poggendorf's Annalen, 1837, p. 184.

‡ Quoted by Pasteur, *opus cit.*, p. 16.

The experiments of Schutze, Schwann, Schræder, and Dusch have been frequently repeated without preventing the growth of fungi.*

Again, it is almost universally considered that the heat of boiling water or cold at zero will destroy all kinds of animal and vegetable life. Indeed, to imagine that the minute molecules or vibrios of which we have been speaking, or small ova and sporules consisting of oleo-albuminous matter without any envelope, would remain in boiling water for hours and retain their vitality, must be regarded as a violent assumption. Three or four minutes' boiling of a hen's egg not only kills it, but converts its whole substance into a hard mass. There is no seed known which, when taken out of its indurated shell or case, is capable of germinating after being boiled for a short time.† Yet nothing is more certain than that long ebullition of various infusions has wholly failed to prevent the formation in them of animal and vegetable growths.

As, therefore, neither calcined air, sulphuric acid, liquor potassæ, gun-cotton, or a boiling temperature have failed to prevent the production of infusoria, or destroy the supposed germs in the air or infusion, I determined, in 1863, to try the effects of all these destructive agents, with the exception of the first, at once, and with the greatest possible care.

On the 17th and 18th of October 1864, and on the 3rd and 13th of October 1865, I performed the following experiments in my laboratory, with the assistance of Dr Argyll Robertson :—

Decoctions of liquorice root, of tea, and of hay were kept at the boiling temperature in a porcelain basin, over a gas flame. Flasks filled with and inverted in the boiling fluid had air pumped into them to the extent of three-fourths of their volume which had passed through (1st) a U-shaped tube containing liquor potassæ; (2d) a hollow glass ball containing gun-cotton; (3d) Liebig bulbs, containing sulphuric acid; and (4th) another U-shaped tube with sulphuric acid. All the bent tubes were filled with fragments of pumice-stone to break up the air, so as to prevent the possibility of any germs passing through in the centre of bubbles. The bent glass tube leading from the

* See Pasteur, *opus cit.*, pp. 34, 35. Pouchet's Hétérogénie, pp. 252, *et seq.*

† See some conclusive experiments recently performed on this subject by Meunier. Comptes Rendus, tom. lxii. p. 992. See also Pouchet's Experiments on the Seeds of Medicago from Brazil. Comptes Rendus, tom. lxii. p. 941.

last U-shaped tube, filled with sulphuric acid and pumice-stone, was also filled with the acid, so as to destroy any germs that might be supposed to adhere to the interior. After the air so prepared had entered the flask, corks, which had been for some time boiled in the infusion, were by means of iron forceps inserted in the necks of the flasks, and the entrance of fresh air prevented. Further, on removing the flask from the boiling infusion, the cork and neck were hermetically closed by plunging them into melted sealing-wax. At the same time bottles or flasks containing the same infusion, but having a similar proportion of ordinary air, were sealed or corked up, so as to be contrasted with the influence of the prepared air.

In another series of experiments, on the 14th of October 1867, with Dr Rutherford, stoppered bottles were used, it having been suggested that sporules or germs might have been concealed in the corks formerly employed although they had been well boiled.

The results were that the infusions in all the flasks in contact with ordinary air were rendered turbid, or covered with fungi in from six to twelve days; whereas all the infusions which were exposed to the prepared air also became turbid and contained fungi, but at periods varying from four to nine months. When the fluid was examined microscopically, bacteria and vibriones were always found. It was also found that rarified air delayed the appearance of these infusoria and of turbidity of the fluid.

It having been asserted by M. Pasteur* that passing air through bent tubes, by hindering the access of germs and allowing them to be deposited on the sides of the glass, prevented the growth of infusoria, twelve bottles were prepared on the 4th and 11th January 1868, four of which contained an infusion of dulcamara, another four a decoction of putrid meat, and a third four, yeast water. The bottles were plunged into, and filled with, the infusion when boiling, inverted in it, and allowed to get cold. The air was pumped gently through a bent tube, five feet in length, having fourteen sharp bends, each four inches long, into three bottles of each series, and ordinary air was admitted to the fourth. The result was, that the fluids in all soon became turbid, so that bent tubes appear to intercept none of the supposed germs. They only delay the result.

* Comptes Rendus, tom. i. p. 306.

In the numerous experiments made by Dr M'Kendrick in 1870, 1871, and 1872, it was found that if the fluid be introduced into a flask, boiled, the neck drawn out, bent so as to form numerous acute angles, and the fluid again boiled, it will remain free from turbidity, or the appearance of bacteria or vibriones for many months, but ultimately the change occurs. The bendings of the tube appear to delay the occurrence of putrefactive changes, but not entirely to prevent it. Moreover, the same effect of delay may be produced by simply having the neck of the tube drawn out to a length of twenty-four or thirty-six inches, without any bend whatever, or by inverting the flask so that the long neck hangs downwards, the atmospheric pressure preventing the escape of the fluid from the flask.

These experiments, on the whole, appear to me to be totally adverse to the atmospheric germ theory, and to indicate that the production or non-production of infusoria depends, for the most part, on the temperature, chemical constitution, density, and other physical properties of the air, rather than on living organisms there, which are developed in the fluid. Still, in every series of experiments, there are one or two exceptions. This has also been observed by Pasteur in most of his experiments, and he attributes them to some currents or limited portions of air being rich in germs, whilst others are free from them.* But that this explanation applies to my laboratory, in which all the experiments described were made, is not probable.

It is now admitted by M. Pasteur that the boiling temperature, that is 100° centigrade, does not prevent the growth of the supposed germs in the atmosphere ; but instead of considering this fact hostile to his theory, he concludes from it that the germs have the power of resisting that amount of heat, and of being most tenacious of life ; but he says, 130° centigrade always destroys their vitality. M. Pouchet, however, has shewn that the air, and the organic matter placed in boiling water, will germinate after they have been exposed to a heat of even 150° centigrade, and he says it may be raised to 200° centigrade, and yet animalcules and fungi will develop themselves.† Dr Bastian has found that even after exposing flasks

* Comptes Rendus, tom. li. pp. 350, 351.

† Ibid. tom. l. p. 1015.

containing organic fluids and hermetically sealed while boiling, to a temperature of 300° centigrade for several hours, animal and vegetable microscopic organisms were found in the fluid.*

In the same manner, air and infusions exposed to intense cold still produce animalcules, but, according to Pasteur, not so readily. Twenty flasks containing boiled infusions, and from which the air was expelled, were opened by him with excessive precaution on the Mer de Glace at Montanvert on the Jura. Notwithstanding the purity and extreme coldness of the air, infusoria appeared in five of his flasks.

As an illustration of the manner in which the controversy on this subject has been carried on in the Academy of Sciences in Paris, I may give a short account of that portion of it referring to the Glacier experiments. M.M. Pouchet, Jolly, and Musset opened eight similar flasks used by M. Pasteur at Montanvert, on the Glacier of the Maladetta, in the Spanish Pyrenees, 9000 feet above the sea, and 3000 feet higher than that of Montanvert, using all the precautions required by M. Pasteur. In addition, before cutting off the ends of their hermetically sealed tubes with a file, previously heated by a lamp, they held the flasks above their heads. Notwithstanding, infusoria appeared in all the infusions a few days afterwards.†

To this communication, presented to the Academy, Sept. 21, 1863, M. Pasteur replies, Nov. 2,‡ saying that he is rejoiced that his learned adversaries have gone to such an altitude to repeat his experiments; but observes that they did not take the necessary precautions. They only had eight flasks, whereas he had twenty; they shook their flasks before opening them, which he took care not to do; and they had the imprudence to use a file, instead of a pair of pincers with long branches, heated in the flame of a lamp. He says that the thumb and fingers holding the file were too near the opening into the flask, and may have conveyed germs there, especially as they were not passed through the flame, as the file was.§ He defies them, if they take sufficient precautions, to obtain infusoria in all their flasks.‖

MM. Jolly and Musset accept the defiance of M. Pasteur,

* Bastian: The Modes of Origin of Lowest Organisms. 1871.
† Comptes Rendus, tom. lvii. p. 558.
‡ Ibid. p. 724.
§ Ibid. p. 725.
‖ Ibid. p. 726.

but every one of his experiments has been repeated by several independent investigators, who have shewn his imagined proofs as to the existence of atmospheric germs to be altogether erroneous. We may conclude, therefore, that living germs are not necessarily the cause of putrefaction and fermentation; neither is it necessary to believe that ferments are living at all—they may be dead. This, if not admitted, seems to be implied by Pasteur himself, who tells us he can now excite these processes, not by fresh yeast only, but by the ashes of yeast.* That they may be induced by dead organic matter which has been subjected to a direct temperature of 150° or 200° centigrade—a heat utterly incompatible with the existence of life—we have seen to have been proved by Pouchet, Jolly, Musset, and others.

The idea that these imaginary germs were the cause of putrefaction, of disease, of blights among vegetables, and other evils, originated with Kircher and the pathologists of the seventeenth century. It has been frequently revived, but always shewn to be erroneous. In 1852, cholera was supposed to be occasioned by a fungus that really existed in the dejections, but which Mr Busk pointed out was the *uredo segetum* of diseased wheat, which entered the body in the form of bread. Certain well-known parasitic diseases are spread by contact, such as scabies, which, as it depends upon an insect burrowing in the skin, may be understood to crawl from one person to another. I succeeded, in 1841, in proving that Favus might be made to grow on diseased surfaces of otherwise healthy persons; but many of our unquestionably infectious diseases, such as smallpox, scarlatina, measles, and typhus, have no such origin. It has been attempted to be shewn, indeed, by Lemaire,† that in the condensed vapours of hospitals and other putrid localities, vibrios may be found; but that vibrios are the cause of these various diseases, is not only not proved, but from what has been stated, is highly improbable. We have previously alluded to the molecules which, according to some, convey some of these poisons. (See p. 104.)

What, then, it may be asked, is the origin of the infusoria,

upon any kind of microscopical inquiry; Messrs Pouchet, Jolly, and Musset, under such circumstances, very properly took no part in the investigation, which, consequently, was altogether one-sided, and of no scientific value.

* Comptes Rendus, tom. lvi. pp. 418, 419. † Ibid. tom. lix. pp. 317-428.

vegetable and animal, that we find in organic fluids during fermentation and putrefaction? In answer to this question, I say they originate in oleo-albuminous molecules, which are formed in organic fluids, and which, floating to the surface, form the pellicle or proligerous matter. There, under the influence of certain conditions, such as temperature, light, chemical exchanges, density, pressure, and composition of atmospheric air, and of the fluid, &c., the molecules, by their coalescence, produce the lower forms of vegetable and animal life.

Other researches may be referred to as confirming these conclusions, such as—

1. *The development of Botrytis Bassiana in Muscardine, a disease of silk worms.*—The true cause of this disease was first ascertained by Bassi in 1835, who shewed that it depended on the presence of a fungus which developed and multiplied within the body of the worm or moth, caused its death, and appeared through the skin in many places as a whitish growth. M. Guérin-Méneville* has observed the development of the fungus filaments from the blood corpuscles of the worm. A similar disease occurs in the house fly in autumn, and is said by Cohn to depend on a mould called *Empusa*, which also originates from the blood cells.†

2. *The development of Penicillium from milk globules.*—This was first described by Turpin in 1837.‡ When a drop of milk is examined after it has acquired an acid reaction, flakes of casein will be found along with bacteria and milk globules variously altered. These milk globules throw off buds from their margin, which grow into mycelium-like filaments, and soon the milk is covered with a whitish mildew, seen with the naked eye.

3. *The development of Bacteria within the laticiferous vessels of plants.*—M. Trecul, a distinguished French botanist, has found numerous minute bodies, of a globular or cylindrical form, some motionless, others shewing slight undulating movements, within the laticiferous vessels of *Apocynum cannabinum*, in the closed medullary cells of the *Ficus carica*, and in the fibre cells of the bark of various plants, such as *Asclepias cornuti*, the common elder, &c. &c.§ He believes these living bodies,

* Comptes Rendus, tom., lvi., p. 574.

† Hedwigia, 1855, p. 59.

‡ Turpin, Ann. des Sc. Nat., 1837 (Zoologie), tom. viii., p. 349.

§ Comptes Rendus (1865), tom. lxi., pp. 158, 432, and 435.

to which he gives the name of *Amylobacteria*, are derived from metamorphosed starchy matter, hence their name.

4. *The occurrence of Bacteria, Fungi, &c., in the centre of the organs of dead animals or in the closed cavities of the body.*—Nothing is more common than to find these organisms in the centre of the brain, in the centre of the liver, even in the hepatic cells, in epithelium cells, or in any part of a dead or even a living body undergoing putrefaction. Berkeley has found a yellow mould within the cerebral cavity of golden pheasants.* Murie has seen fungus-growths within the abdomino-pleural membrane of a kittiwake gull, of a great white-crested cockatoo, and of a rough-legged buzzard.† Bacteria and fungi have often been found in eggs. Helmarecht found spores in the aqueous humour of the human eye.‡ Numerous other examples might be given, but enough has been brought forward to shew that foreign living organisms have originated in situations entirely removed from the air.

ABNORMAL REPRODUCTION.

This consists in the various alterations which may occur in the different stages of the generative functions, and include,—1st, Diseases which arrest or modify ovulation ; 2d, Diseases, nutritive or nervous, which impede fecundation, and occasion barrenness in the female, or impotence in the male ; 3d, Diseases of the embryo, causing various kinds of monsters, from arrest or excess of development in one or more of its parts. This last subject is now generally studied under the name of *teratology* (τέρας, *monster*), and has in recent times become a very extensive one. Congenital malformations of the fœtus were formerly considered as indicative of some misfortune,—as the effect of witchcraft, or as offsprings of the evil spirit. They are now not only recognised to originate in natural derangements of embryonal development, but the laws which govern such derangements have to a great extent been determined. From these it has become evident that monstrosities are not the result of chance, but are always induced by alterations in the known processes which regulate reproduction, and the evolution of the ovum and its contents. Hence in this, as in every other disordered condition, the real source of the abnormality is to be sought for,

* Berkeley, Introduction to Cryptogamic Botany, p. 260.

† Murie, Report of British Association, 1871.

‡ Robin's "Végétaux Parasites," 1853, p 370.

not only in the investigation of that condition itself, but in the knowledge, first, of the healthy or physiological state ; and secondly, of the manner in which it has become deranged. In all our inquiries, it must be apparent that disease is morbid physiology ; and such is the aspect in which we have endeavoured to place it before the reader.

ON DEATH.

Death is the permanent cessation of those properties and functions which constitute life. In this wide sense, it must be apparent that the textures are continually dying, in the same manner that they are continually being generated. What we have described as the secondary digestion essentially consists in the removal of the particles of the body which have been worn out,—fulfilled their functions, and died. Thus, death is molecular, cellular, fibrous, or tubular, in proportion as these various organic elements become degenerated, and disappear to make way for others which enjoy activity or life, and in their turn die, enter into new chemical combinations, and are excreted like their predecessors. In the more common acceptation of the term, however, death may be considered as *partial* or *general*. Partial death of the animal body is caused by those diseases or injuries which produce mortification and ulceration in soft, and necrosis and caries in the hard parts, to a greater or less extent. Of this we have already spoken, and therefore need only treat of general death of the system. This has been variously considered as *natural* or *unnatural ;* by the former meaning death from old age or gradual decay, and by the latter, death from diseases or violence. In this latter case, death may be gradual or sudden, and be induced by a great variety of agents. It may be said, however, that all the modes of death are reducible to three, viz. : 1st, Death by syncope—that is, beginning at the heart ; 2d, Death by asphyxia, beginning at the lungs ; and 3d, Death by coma, beginning at the brain.

Death by syncope.—All causes which arrest the action of the heart occasion stoppage of the circulation ; a circumstance which interferes with the due performance of the vital functions ; and death is the consequence. It may occur through the nervous system, through feebleness of the muscular walls of the heart itself, or through loss of blood. As examples of the first

method of causing syncope, may be cited concussion, or all sudden shocks to the system—as from violent blows or injuries, extensive lesions, violent mental emotions, a stroke of lightning, exposure to the sun (or *coup de soleil*), and certain poisons which, acting especially on nerves going to the heart, paralyse its rhythmical motions, as aconite, digitalis, &c. Syncope, from feebleness of the muscular walls is illustrated from the effects of long-continued violent exertion, starvation, and disease of its textures, especially that now recognised as fatty degeneration, one of the most common causes of sudden death. Lastly, excessive loss of blood, whether from direct external injury to a large vessel, sudden bursting of an internal vascular tumour or aneurism, disease of the coats of an artery or vein leading to sudden or to long-continued loss of blood, are among the frequent causes of syncope.

Death by asphyxia.—This is produced by all causes which interrupt the act of respiration, or the access of oxygen, so necessary for carrying on the nutritive functions, and has been previously referred to (p. 233). It is now ascertained that mere obstruction of air does not immediately act upon the heart, which not only continues to contract for a time, but even sends venous blood through the arterial system. From the numerous investigations which have been made to determine in what manner the vital actions are arrested in asphyxia, it would appear that at first non-aërated or venous blood passes freely through the lungs to the heart, from whence it goes to all parts of the system. It operates on the brain, however, as a poison, rapidly suspending the sensorial functions. The capillaries of the lung next refuse to transmit non-oxygenated blood, in consequence of which it is not returned to the right side of the heart, and thus the vital actions cease. These effects are produced with greater or less rapidity, according as the occlusion of air is more perfect, as in cases of drowning and strangulation. In diseases of the heart and lungs, the same results are produced more slowly. The only poisons which operate upon the lungs directly causing asphyxia are certain so-called poisonous gases, such as carbonic acid gas, the fatal effects of which, however, are not so much to be ascribed to any noxious properties it possesses as to the absence of free oxygen.

Death by coma.—This is caused by all circumstances which suspend the sensorial functions by first operating on the brain.

We observe it produced from the long-continued action of cold, from the influence of narcotic poisons, especially opium and chloroform; and from such injuries of the brain, from without or within, as are not necessarily connected with shock. If a violent blow be given to the head of an animal, it may be observed to suffer from shock or syncope; the heart flutters, and the pulse is weak. But if it recover from this, the heart's action may be restored, while sensation is suspended, and it dies comatose. If shock be avoided during the operation, the brain of an animal may be removed, producing coma or stupefaction, which will ultimately kill, although for some time the circulation and respiration continue. In apoplexies, fevers, and other diseases, similar effects are observable.

It should not be overlooked that death in many cases is produced by a conjunction, or by the rapidly-following results of two or all three of these modes. Thus, chloroform may kill from the conjoined stupefying action on the brain, as well as from difficulty of respiration. Coma, from pressure on the brain, may, by influencing the *medulla oblongata*, affect the pneumo-gastric nerves, which send branches to the heart and lungs. In this case, death is the most rapid—occurring in all three ways. Hence the humane effort of the hangman not only to produce strangulation, but by dislocation of the bones of the neck, to crush the upper part of the spinal cord.

The preceding observations evidently indicate that, in our endeavours to produce recovery from either of these states, much will depend upon the correct information we derive as to the causes producing them. In syncope, our efforts will be directed to restore the action of the heart by stimuli, a proper position, checking hæmorrhage, &c.; in asphyxia, to reproduce respiration; and in coma, to remove any cause which, by pressure on the brain from without or within, interferes with its functions.

PART III.

PRACTICAL PHYSIOLOGY.

By the term "practical," is not understood giving lectures on practical as distinguished from theoretical subjects. What I understand by it is causing the student himself to perform with appropriate instruments the necessary investigations, so that he may learn the art of observation, and obtain the necessary manual dexterity for arriving at exact results. Thus, practical chemical physiology consists not in being shewn by the teacher how to analyse the various fluids and solids of the animal frame, but in his doing this himself. Practical histological physiology is not merely examining objects and preparations, but in causing the student to manipulate the microscope, demonstrate for himself, and describe what he sees. Practical experimental physiology, in like manner, consists not only in witnessing, but taking part in, the performance of experiments on animals, with all the modern instruments of precision that science in recent times has placed in our hands.

PRACTICAL PHYSIOLOGICAL CHEMISTRY.

Practical physiological chemistry includes an examination of the solids and fluids of the body. The physical properties of the substance, such as form, colour, hardness, specific gravity,

&c., must first be accurately observed. It is then subjected to chemical analysis, either quantitative or qualitative, and a systematic mode of procedure followed.

I. *GENERAL QUALITATIVE EXAMINATION OF AN ANIMAL FLUID.*

The examination should be made in the following order :—

1. *Reaction to test paper.*—The tests used are blue litmus paper, which becomes red when dipped in an acid fluid, and red litmus, which turns blue, or yellow turmeric paper, which becomes brown when immersed in an alkaline fluid. An acid reaction of a liquid indicates the presence of free acids or of acid salts, whereas an alkaline reaction is produced by free alkalies, alkaline phosphates, or carbonates. If an alkaline reaction disappear on gently heating the paper over the flame of a spirit lamp, it informs us that the alkali is volatile ; if it remain, the alkali is fixed.

2. *Filter, or strain.*—A fine, white, thin blotting paper is the best filtering medium for ordinary purposes. When the liquid passes through the paper turbid, it should be returned once or twice to the filter till it comes through clear. The precipitate should be preserved by being scraped off with a spatula of ivory, platinum, or steel, or it may be washed off with a gentle stream of water from an ordinary wash bottle. Sometimes it is necessary to strain through one or several folds of muslin or cotton.

3. *Heat a portion of the filtrate.*—This should be done in a test tube over a spirit lamp. If no precipitate is formed, albumin is absent. If a precipitate appear, it may be albumin or phosphates. Add a few drops of dilute hydrochloric or nitric acid. If the precipitate disappear, *albumin* is absent (p. 8), but *earthy phosphates* may be there (p. 28). These must be looked for and isolated by special tests.

4. *Add to the fluid a solution of Ferrocyanide of Potassium.*—If there be no precipitate, casein and globulin are absent. If a precipitate fall, take a fresh portion of fluid and divide into two parts. To one add a solution of chloride of calcium and apply heat. A precipitate indicates *casein* (p. 9). Carefully neutralise the other portion,—if acid, with an alkali, if alkaline, with a few drops of acid,—and observe whether a precipitate be

formed at the point of neutralisation. If a precipitate appear, *globulin* is present (p. 10).

5. *To a portion of the liquid add acetic acid.*—A precipitate may be formed. If so, it is chondrin, mucus, or pyin. To a portion add a solution of common alum or sulphate of copper. If a precipitate fall, soluble in excess of the reagent, *chondrin* is present (p. 11). To another part add a solution of corrosive sublimate, and if there be a precipitate, we have pyin, or mucin (p. 11). Neutral acetate of lead distinguishes the two, giving a copious precipitate with *pyin* and a very slight turbidity, or none at all, with *mucin*.

6. *Evaporate a few ounces of the liquid to one-sixth of its bulk.*—If a jelly form on cooling, we have *chondrin*, or *gelatin* (p. 10). For the mode of distinguishing chondrin see last paragraph. If there be a precipitate, it may be *urates, phosphates, sulphates, allantoin, tyrosin, hippurate of calcium*, or *benzoic acid.* These must be examined microscopically, and by special tests, to be afterwards described.

7. *Evaporate a few ounces to a syrup, and allow it to stand for forty-eight hours.*—If crystals form, allow the fluid to stand as long as they increase. Such crystals may be *creatin* (p. 18), *creatinin* (p. 18), *leucin* (p. 17), *allantoin* (p. 16), *taurin* (p. 16), *sarcin* (p. 16), *inosite* (p. 27), *hippurates of potash or soda* (p. 15), *sodium chloride*, and other salts (p. 28). The next point to determine is, do the crystals consist of organic or inorganic matter. Heat a small portion on a clean bit of platinum foil. If it blacken when strongly heated, organic matter is present. If a whitish-coloured residue be left, after heating strongly, it consists of inorganic matter ; and the probability is we have an organic acid united with an inorganic base. If the crystals consist solely of organic matter, they must be examined by processes of organic analysis for nitrogen, sulphur, and phosphorus, and their chemical composition determined. The tests for these substances will be given when we treat of special fluids and solids.

8. *Separate the crystals from the syrup, and exhaust the residue with alcohol of specific gravity* 0·833.—Treat as follows :

DIVIDE ALCOHOLIC SOLUTION INTO SIX PORTIONS.

1. Concentrate, dilute with water, place a few drops on a porcelain plate, and add a drop of nitric acid. Play of colours indicates . . . . . *Bile pigment* (p. 32).

2. Concentrate, dilute with water, place in a test tube, add ½ bulk of syrup, then allow a drop or two of sulphuric acid to flow down side of test tube. Play of colours at junction of fluid and syrup indicates *Bile acids* (p. 12).

3. Evaporate to dryness, dissolve in water. Apply the tests for sugar according to methods to be afterwards described . . . . . *Sugar* (p. 25).

4. Evaporate to a small bulk, add nitric acid; laminar crystals of nitrate of urea separate out—indicating *Urea* (p. 14).

5. Mix with a very strong solution of chloride of zinc. A crystalline precipitate indicates . . . *Creatin and Creatinin* (p. 18).

6. Heat with oxide of zinc, filter while hot, and evaporate a drop on a glass slide. Club-shaped crystals of lactate of zinc indicate . . . . . *Lactic Acid* (p. 27).

9. *Evaporate part of original fluid to dryness, pound the residue in a mortar, and exhaust with ether.*—This etherial extract containing fats in solution, is evaporated and further examined.

10. *Incineration.*—The residue insoluble in ether is incinerated in a platinum capsule, and the ash examined by the ordinary methods of inorganic analysis.

General Conclusion.—By these ten processes we ascertain whether the fluid be acid, alkaline, or neutral, and whether it contain albuminates, albuminoids, or albuminous derivatives. We also obtain a knowledge of the presence of fatty matters or mineral principles.

II. *QUALITATIVE AND QUANTITATIVE ANALYSIS OF SPECIAL ANIMAL FLUIDS.*

The fluids we specially examine are—(1.) The *blood;* (2.) The *chyle;* (3.) The *lymph;* (4.) The *saliva;* (5.) The *gastric juice;* (6.) The *pancreatic juice;* (7.) The *bile;* (8.) The *urine;* (9.) The *sweat.* Under this head we may also describe the analysis of (10.) The *fæces.*

Analysis of Blood.

For the chemical constitution of the blood, see pp. 239, 240. The analysis is conducted in the following manner. The reaction is usually slightly alkaline.

1. *Water.*—Weigh out a certain definite quantity, evaporate in a porcelain basin over a water bath, and dry the residue in a hot air chamber, at a temperature between 120° and 130° C. Weigh again. The loss in weight will represent the water.

2. *Fibrin.*—Receive an ounce or two of the blood into a glass vessel as it flows from a vein, stir it up for ten minutes with a glass rod or twig of birch till the fibrin is separated. The blood, with the fibrin, is then weighed, and strained through muslin to separate the fibrin, which is well washed with water, dried, and boiled with alcohol and ether to free it from fat. The alcohol and ether are driven off by evaporation, the fibrin is dried at 120° C., and again weighed. The weight indicates the amount of fibrin in the portion of blood examined.

3. *Albumin and substances coagulated by heat.*—A few ounces by weight of blood are acidulated with acetic acid, and added drop by drop to boiling water. The aqueous liquid is then poured upon a carefully weighed filter, and the coagulum separated. The coagulum is then washed on the filter with boiling water, and dried at 120° C. It is then weighed, the weight of the fibrin deducted, and the balance represents albumin, and a small amount of certain other substances coagulable by heat.

4. *Fat.*—A weighed quantity of blood is dried at 100° C., and treated with ether. The etherial solution is filtered into a platinum capsule, in which it is slowly evaporated, and the residue dried at 100°. The weight of residue gives the amount of fatty matter.

5. *Extractive matter and mineral constituents.*—The filtrate obtained in the estimation of the albumin is evaporated on a water bath, and the residue dried and weighed. It is then burnt at as low a temperature as possible. An ash is left, which consists of mineral constituents. The weight of this ash is deducted from the weight of the dried residue, and the difference is the extractive matter. The mineral matter may also be obtained by the process of dialysis (p. 118).

6. *Mineral matter.*—A quantity of the blood is weighed, mixed with ignited carbonate of soda, dried and burnt at as low a temperature as possible. The residue is the ash. The incineration must be done with great care, as the chlorides of potassium and sodium volatilise at a high temperature ; phosphates may be decomposed, and sulphates reduced to sulphides. The ash is then treated as follows :

DIVIDE INTO TWO PARTS.

First portion—Treat with dilute nitric acid; add nitrate of silver to precipitate the chlorine as chloride of silver	*Chlorides.*
Second portion—Treat with dilute hydrochloric acid. Divide into three portions.	
1. Add chloride of barium to precipitate sulphuric acid as sulphate of barium . . .	*Sulphuric acid.*
2. Mix with ammonia and a little acetic acid—	
(1.) Add oxalate of ammonia to throw down lime as oxalate of lime . . .	*Lime.*
(2.) Add excess of ammonia to throw down all the magnesia and part of the phosphoric acid	*Magnesia and phosphoric acid.*
(3.) Add sulphate of magnesia to throw down the rest of the phosphoric acid . .	*Phosphoric acid.*
3. Add a little oxalic acid to throw down the lime, and also a few drops of ammonia, and phosphate of ammonia, to throw down the magnesia. Filter. Dry the residue, and dissolve in hydrochloric acid. Add tetrachloride of platinum, which throws down all the potassium as a yellow crystalline precipitate. In the fluid, on evaporation, will be found the sodium salt .	*Potassium and Sodium.*

7. *Serum and coagulum.*—A certain amount of blood is allowed to stand in a vessel till all the clot has separated. The clot is then carefully detached by a needle or sharp knife from the side of the vessel. The blood is weighed, and after the clot has contracted as much as possible, the serum is poured off. The clot is dried by means of blotting paper and again weighed. If we deduct the weight of the clot from the total weight of the blood, we find the proportion of serum.

8. *The colouring matter of the blood, Hæmoglobin, Hæmataglobin, or Hæmatocrystallin.*—(See p. 31, and Plate I. figs. 20 and 22.) Take a few ounces of blood, beat with a twig of birch for a quarter of an hour to separate the fibrin, strain through a linen cloth along with a mixture consisting of one volume of a saturated solution of sodium chloride and nine volumes of distilled water. A precipitate is formed, which is shaken up with a little water and from four to ten times its volume of ether.

The ether is drawn off with a pipette after several hours, and is found to contain cholestrin. An aqueous solution of the residue is now brought to a low temperature, and a pulpy mass forms consisting of crystals of hæmoglobin.

9. *Estimation of Iron.*—This may be readily done by burning the hæmoglobin obtained by the process just described. When hæmoglobin is burnt, a small residue of ferric oxide is left, from the weight of which the amount of iron is then calculated (every 100 parts of ferric oxide containing 70 parts of iron).

10. *The optical properties of Hœmoglobin.*—These may now be examined according to the method and optical principles described at pp. 32 and 138. For this purpose we require an instrument termed a *spectroscope*, of which there are two principal varieties, the ordinary spectroscope and the micro-spectroscope. When there is a large quantity of blood available, as in most physiological experiments, the ordinary spectroscope should be employed, but when only a drop or two of blood can be obtained, we use a spectrum apparatus fitted to a microscope. The construction of a spectroscope depends on the optical principle that when a ray of sunlight, or a ray of white light from an artificial source, passes through a prism it splits up into a number of individual coloured rays, in the following order, running from left to right—red, orange, yellow, green, blue, indigo, purple, and violet (p. 138). The ordinary instrument consists essentially of three parts : 1st, a tube having at the end an adjustable slit for admitting the light ; 2d, a prism made of flint glass, or a triangular glass bottle filled with Disulphide of Carbon ; and 3d, a magnifying glass or telescope for increasing the apparent size of the spectrum. The tube of the telescope must be placed in a different direction to the tube carrying the slit, because the coloured rays forming the spectrum form an angle with the incident rays as they enter the prism.* When a thin layer of a liquid, such as arterial blood (which contains oxy-hæmoglobin, p. 32), is placed in front of the slit, the colouring matter intercepts certain coloured rays of the spectrum, so that two dark bands are seen between the yellow and green of the ordinary spectrum. For this experiment the blood must be greatly diluted with water and a small test tube, or still better, a vessel having parallel sides of thin glass, filled with the solution, is placed before the slit. To increase the amount of light, it

* Schellen, Spectrum Analysis. London, 1872.

may be concentrated on the slit by means of a powerful bull's-eye condenser.

The microspectroscope of Sorby and Browning is a spectrum arrangement which can be applied to any microscope by fixing it in the place of the ordinary eyepiece, so that spectroscopic investigation of an object can be pursued without any change in the manner of using the instrument. By means of this instrument the two absorption bands of arterial, and the single one of venous blood, can be readily recognised.

Recently, W. Preyer* has applied the method of spectrum analysis to the determination of the quantity of hæmoglobin. "The determination depends upon the fact that a concentrated solution of hæmoglobin in a layer of certain thickness is opaque, even in strong illumination, to all rays except the red, whereas less concentrated solutions in a layer of the same thickness give passage to other rays besides the red and orange, and especially to a portion of the green. If, therefore, a measured quantity of blood placed before the slit of the spectral apparatus be diluted with water till green light appears in the spectrum, and if the proportion of hæmoglobin in a solution which transmits the green under exactly similar circumstances has once for all been determined, it is easy to estimate the percentage of hæmoglobin in the blood under examination."†

11. *Effect of passing a stream of carbon monoxide through blood.*—When a stream of carbon monoxide is passed for some time through blood, and a little of this blood is examined by the spectroscope, the two absorption bands of oxy-hæmoglobin will be seen. Hoppe-Seyler of Strasburg has pointed out that these absorption bands do not disappear and give place to a single band on the addition of sulphide of ammonium even after several days, and he proposes this unalterability of blood containing carbon monoxide by ammonium sulphide as a test for the presence of this gas in the blood. ‡

12. *Detection of blood stains by the formation of Hæmin crystals.* §—Sprinkle a few grains of common salt on the stain. After a few minutes add a few drops of glacial acetic acid.

* Ann. Chem. Pharm. cxl., p. 187.
† Watt's Dictionary of Chemistry. Supplement, p. 353.
‡ Hoppe-Seyler, Zeitschrift Anal. Chem. iii. 439. Jahresb. 1865. p. 745.
§ Brücke, Jahresb. 1857. P. 609.

Scrape off as much matter as possible from the stain, and place it on a glass-slide. If necessary to moisten still more, add another drop or two of glacial acetic acid. Then carefully put on a thin covering glass, and lay the slide aside for two hours in a moderately warm place, say four feet from an ordinary fire. At the end of this time, examine with a power of 250 diameters and the black crystals of hæmin will be found. (Plate I. figs. 19 and 21.)

13. *Guiacum test for blood.*—Another test for blood is given by tincture of guiacum. A drop of freshly prepared tincture is placed on a bit of white blotting-paper. A drop of any solution suspected of containing blood is added, and afterwards a drop or two of hydrogen dioxide (H_2O_2). If blood be present, the stain on the blotting-paper soon turns from green to blue. This, however, is not a test to be absolutely depended on, because saliva, gum arabic, citrate of iron, &c., give the same reaction. As a negative test, however, it is valuable, because if the blue colour be not obtained, blood is certainly not present.*

14. *Gases of the blood.*—The method of L. Meyer for determining the gases of the blood is as follows :—The blood is diluted with ten times its bulk of water, and the gases are collected by boiling the liquid in vacuo at a gentle heat. The *free* gases are thus obtained. A few crystals of tartaric acid are then added, the blood is again boiled, and the *combined* gas is liberated. The gases of the blood consist of oxygen, nitrogen, and carbonic acid. They are introduced into a special apparatus for the analysis of gases. † The carbonic acid is absorbed by caustic potash, and the amount thus determined. The amount of oxygen is found by exploding the mixture with an excess of hydrogen, and one-third of the total amount of contraction caused by the explosion is the quantity of oxygen present. To ascertain the nitrogen, all the other gases must be removed, and the residue consists of nitrogen. The gases may also be collected by means of an air pump, of which the best form for this purpose is the mercurial air pump of Sprengel.‡

* Van Deen, Zeitschrift Anal. Chem. ii., p. 459. Taylor, Guy's Hospital Reports. 1868.

† For details as to analysis of gases, see Watt's Dictionary of Chemistry, vol. i. p. 268.

‡ Quicksilber—Luftpumpen ; Müller—Pouillet's Lehrbuch der Physik. I. Bd. s. 211 ; II. Bd. s. 941. Also Dr Wüllner's Lehrbuch der Experimentalphysik. I. Bd. p. 365.

The serum of blood has also been found to contain sugar, urea, uric acid, creatine, creatinine, hippuric acid, hypoxanthin, leucin, and tyrosin, which substances exist only occasionally in very small quantity, and are difficult to isolate.

Analysis of Chyle.

This fluid must be obtained from the thoracic duct of an animal killed during digestion. It is an opalescent, milky liquid, having a saline taste and a very weak alkaline reaction. The specific gravity varies from 1012 to 1025. It may be analysed by pursuing the same method described for the blood. Numerous analyses made by Pelouge, Frémy, Rees, Simon, and Nasse vary as to the relative proportions of its constituents.

Analysis of Lymph.

When obtained fresh from the lymphatic vessels, its reaction is usually alkaline. It soon coagulates, forming a jelly-like coagulum. The coagulum will be found to consist of a substance identical with fibrin. The fluid contains albumin, which may be readily separated by dropping into boiling water. A small amount of fat may be found by shaking up with ether. A very small amount of extractive matter may be taken up with alcohol. The salts are separated either by obtaining the ash, or by means of dialysis. They are found to consist of sodium chloride, alkaline carbonates, salts of ammonia, sulphates, and phosphates.

Analysis of Saliva.

This may be obtained by introducing a small silver canula into either Stenson's or Wharton's duct, and may be examined according to the method already detailed (p. 445). The reaction is alkaline. Evaporation will yield the amount of solid residue. On allowing saliva to stand for some time in a beaker, it becomes opalescent or turbid, and bubbles of gas form on the surface. This is due to a deposition of calcium carbonate and the escape of carbonic acid. When a drop of saliva is placed on a porcelain lid, and a drop of Ferric chloride added, a blood red or yellowish red colour is obtained owing to the presence in saliva of sulphocyanide of potassium.

Separation of ptyalin.—Dilute the saliva with dilute phosphoric acid, add lime water, and thus obtain a precipitate containing phosphate of lime, ptyalin, and a small amount of

albumin. Shake up with distilled water. The ptyalin is dissolved and separated from the other two substances. This solution of ptyalin may be concentrated by evaporation, and its action on starch observed.

Analysis of Gastric Juice.

This fluid may be collected by making in a dog a gastric fistula, into which a silver or gold canula, having a stop-cock, is introduced. The gastric juice is transparent, colourless, or slightly yellowish. It has a sourish odour. When obtained during fasting it is neutral or slightly alkaline, consisting chiefly of mucus, but during the ingestion of food it is strongly acid (p. 203).

Water.—This may be calculated by evaporating a given weight of gastric juice and weighing the residue. Deduct this weight from that of the gastric juice, and the difference yields the weight of water.

Preparation of pepsin.—Add alcohol, which precipitates the pepsin. Evaporate and pepsin is obtained, the properties of which should be examined as follows:—Dissolve in warm water. Divide into five portions, and treat as follows:

1. Add solution of corrosive sublimate.
2. Add protochloride of tin.
3. Add basic acetate of lead.
4. Add tannic acid.

} *Dense precipitate of pepsin.*

5. Add a few drops of hydrochloric or lactic acids, then several pieces of minced meat, and lay aside in a warm place (100° F.) for four hours. *The meat will then be found soft, whitish in colour, and partially digested.*

Acid of the juice.—Great differences of opinion have prevailed as to this point, Bernard * and others asserting that no hydrochloric acid is present, but only lactic acid; while Bidder and Schmidt † declare they have found free hydrochloric acid. Gastric juice will always be found to give a precipitate with nitrate of silver or oxalic acid. It is probable that sometimes one sometimes the other acid is present, while occasionally both may be there.

Ash of the juice.—When examined in the usual way, sodium

* Bernard, Leçons de Physiologie Expérimentale. Paris, 1856. ii.

† Bidder und Schmidt, Die Verdauungssäfte und der Stoffwechsel. Mittau und Leipsig, 1852.

and calcium chlorides; potassium, sodium, and calcium sulphates; and calcium carbonate and phosphate, will usually be found.

Analysis of Pancreatic Juice.

This fluid may be obtained in sufficient quantity for analysis by cutting the duct, inserting a canula, and collecting the juice in a caouchouc bag. It is clear, viscid, and has an alkaline reaction (p. 203). The amount of solid matter may be ascertained by evaporating a known quantity, drying carefully in a hot-air chamber over sulphuric acid and under the receiver of an air pump, and weighing the residue.

Preparation of pancreatin.—Add to the juice its own bulk of alcohol. A white flaky precipitate falls. Filter, dry carefully, and dissolve the filtrate in water. This is probably a solution of pancreatin along with a protein substance allied to casein. Danilewsky* has tried to shew that in the pancreatic juice we have three ferments: one which acts on starch and albumin and fats; a second, which acts on starch and albumin but not on fats; and a third, which acts on starch but not on albumin.

Analysis of Bile.

Ox-bile is usually examined because easily obtained. It is a transparent greenish liquid, having a ropy character due to admixture with mucus. This last-named characteristic may be demonstrated by pouring the bile from one vessel to another. *Sp. gravity* about 1002. *Reaction* slightly alkaline, or neutral (p. 203). The analysis of bile may be conducted as follows †:—

1. *Mucus.*—Precipitate the mucus by adding to the bile half its bulk of alcohol (83 per cent); filter, wash the precipitate with spirit, afterwards with water, dry in a hot-air chamber, and weigh.

2. *Solid matter.*—Evaporate the fluid obtained by the last operation (which is bile free from mucus) first over a water bath, then under the air pump on a sand bath heated to 100° C. Cool in vacuo, after which allow dry air to pass into the receiver, weigh quickly, and the result will indicate the amount of solid matter. The air may be dried before passing into the receiver by passing it over chloride of calcium.

3. *Fat and cholestrin.*—Pour upon the solid matter obtained

* Virchow's Archives, xxv. p. 279

† Watt's Dictionary of Chemistry, article—Bile, p. 585.

as above, a few ounces of ether, and allow it to digest for twenty-four hours. Thus an etherial extract of the fat and cholestrin will be obtained, and the amount of these substances can be determined by evaporation and weighing (p. 19). Cholestrin is easily prepared by boiling a little powdered gall stone in alcohol along with a few drops of caustic potash to dissolve fatty matters. From this boiling solution cholestrin separates out in laminæ, often having a small notch at one corner (Plate I. fig. 19).

4. *Bile acids.*—These are taurocholic and glycocholic acids (see p. 12), united with sodium and potassium. They may be obtained by either of two methods.

First method.—After the third operation of removing the cholestrin and fat by ether, an insoluble residue will remain. This must now be treated with cold absolute alcohol, which dissolves the alkaline salts of the bile acids along with a small proportion of pigment. Evaporate the most of the alcohol, add ether to the concentrated alcoholic solution, and set the liquid aside for forty-eight hours in a cool place. A precipitate is thus formed of the bile-acid-salts. Filter, and dry and weigh the precipitate. To estimate the amount of alkali united with the acids, add to the ether precipitate a little sulphuric acid. Thus sulphates of soda and potash will be formed, which may be separated, weighed, and the amount of the alkalies calculated.

Second method.—Add to the bile along with alcohol, basic acetate of lead; filter, wash the precipitate with a solution of carbonate of soda; evaporate to dryness. We thus obtain the sodium salts of the bile acids, to which we add first a little absolute alcohol, and afterwards dilute with water.

The chemical composition of the bile acids has been already given at pp. 12 and 16. It is to be observed that the sulphur of the bile exists in taurin, one of the ingredients of taurocholic acid. The relative quantities of the two acids may therefore be determined by finding the amount of sulphur in the ether precipitate; every six parts of sulphur correspond to 100 parts of taurocholate of sodium.*

5. *The residue*, which is usually very small in amount, contains pigment, alkaline and earthy phosphates, chloride and carbonate of sodium. The amount of inorganic salts may be

* For the mode of estimating the sulphur see Watt's Dictionary of Chemistry, article—Analysis (organic). Estimation of Sulphur, p. 247.

determined by incineration and subsequent chemical examination.

Bile pigments.—These are five in number (p. 32), but they may be conveniently divided into the brown and the green.

The brown pigments are soluble in chloroform, while the green are not, and thus we have a ready means of separating the two. The *brown* pigments are bilirubin (red) and cholophæin (brown). To distinguish the cholophæin from the bilirubin, according to Brücke,* evaporate the chloroform solution, wash the residue with alcohol and ether, until a brick-red powder, soluble in ammonia, is obtained (bilirubin); add to the ammoniacal solution a little hydrochloric acid, and cholophæin will be precipitated as yellowish-brown flakes.

The *green* pigments are represented chiefly by biliverdin, which is insoluble in chloroform, but easily soluble in alcohol, benzol, or disulphide of carbon. The green pigment may be formed from the red pigment by boiling an alkaline solution of the latter; and, according to Städeler, the change may be represented by the following equation :—

$$\underset{\text{Bilirubin.}}{C_{16}H_{18}N_2O_3} + H_2O + O = \underset{\text{Biliverdin.}}{C_{16}H_{20}N_2O_5}$$

The chemical reactions of alkaline solutions of these pigments may be examined as follows :—

Reagent.	Bilirubin.	Biliverdin.
Chloroform.	Soluble.	Insoluble.
Barium chloride.	Precipitate.	No precipitate.
Calcium chloride.	Precipitate.	No precipitate.
Neutral lead acetate.	Red precipitate.	Dark green precipitate.
Silver nitrate.	Red-brown precipitate.	Dark green precipitate.
Nitric acid.	Play of colours ending in a green.	Play of colours ending in a yellow.

Optical properties of bile acids and pigments.—According to Hoppe-Seyler,† the bile acids rotate the ray of polarized light to the right. The highest rotatory power is shewn by cholic acid (p. 16). An alcoholic solution of the bile pigments when examined, by the spectroscope, give absorption bands in the vicinity of the letters C and D of the spectrum. Biliverdin absorbs light at both ends of the spectrum, and if not much diluted transmits

* Brücke, J. pr. Chem. lxxvii. 72. Jahresb, 1859, p. 637.

† Hoppe-Seyler, J. pr. Chem. lxxxix. 257. Bull. Soc. Chim. v. 622.

only green light. A very weak solution absorbs only the extreme red. Numerous modifications of these absorption bands may be obtained by acting on the pigment solution with nitric or hydrochloric acids, or by lead acetate or calcium chloride.*

Tests for bile, bile acids, and bile pigment.—It is often of importance to ascertain the presence of bile in urine or other fluid. For that purpose the following tests may prove serviceable:—

1. *Noel's test for bile.*—Immerse a strip of blotting paper for a few minutes in the fluid, dry, and add a drop of nitric acid containing a little nitrous acid. If bile be present, it will assume a violet colour, changing to red or yellow.†

2. *Pettenkofer's test for bile acids.*—To a little diluted bile, or any liquid containing bile, in a test tube, add a little powdered white sugar, or its equivalent of syrup. Then pour in of strong sulphuric acid (very gradually) rather more than half the bulk of the liquid. By this means the temperature is gradually raised to the proper point, and a deep purplish-crimson colour makes its appearance. This test frequently fails when applied to urine, but if an attempt is made to separate the bile acids by the second method above described, and the test applied to the alcoholic and aqueous solution of the acids, very minute quantities will give the reaction.

3. *The nitric acid test for the bile pigments.*—Place a drop of the suspected fluid on a white porcelain plate, add carefully a drop or two of strong nitric acid, and at the point of contact of the fluid with the acid there will be a play of colours, passing through a red, green, pink, blue, violet, and yellow. The appearance of the green colour, though often evanescent, is indicative of bile. A play of colours may be obtained by the action of nitric acid on the pigment in concentrated urine, but it never shews a green tinge unless bile is present.

4. *The silver oxide test for bile pigments.*—Boil the fluid with an ammoniacal solution of silver oxide. Acidulate the filtrate with a few drops of hydrochloric acid. A purple colour will be produced if biliverdin be present, owing to the formation of an artificial compound called bilipurpin.

Biliary calculi or *gallstones.* — These concretions consist usually of a nucleus of mucus or inspissated bile, which becomes coated with cholestrin. Upon this successive layers of earthy

* Jaffe, Zeitschrift f. Chem. [2] v. 666. Maly, ibid. [2] v. 365.

† Noel, J. Pharm. [3] xli. 354.

phosphates and carbonates are deposited, often tinged with the bile colouring matter.

Analysis of the Urine.

This secretion, from its great clinical importance, requires to be carefully examined. Urine may be either healthy or abnormal, that is to say, it may contain normal constituents, such as water, inorganic salts, and organic substances ; or it may contain occasional or abnormal constituents such as blood, albumin, sugar, fat, &c.

1. *Physical properties.*—In its fresh state it is clear, and of a light-yellow colour, has a peculiar odour, a bitter taste, and an acid reaction. With regard to colour, Vogel has classified the numerous varieties of shades of colour we constantly meet with into three groups : 1. Yellow urines ; 2. reddish urines ; 3. brown or dark urines. These again may be subdivided. The varieties of colour depend not on different pigments, but on variations in the quantity of the same pigment. This may be readily shewn by evaporating a light-coloured urine. We find that as the fluid diminishes in quantity, the colour becomes darker. On the other hand, when we dilute any dark-coloured urine, the colour becomes much lighter. The colouring matter of the urine, so far as known, has been already described at p. 34.

The *odour* of urine is affected by food, or medicine, for instance, asparagus, turpentine, saffron, cubebs, &c., may be detected. Turpentine gives urine the odour of violets.

2. *Reaction.*—The urine of carnivorous animals is acid, except during digestion, while that of herbivora is alkaline, except after a prolonged abstinence from food. The cause of the constantly acid reaction of healthy human urine has been disputed. Liebig's view is, that it depends chiefly upon the presence of an acid phosphate, such as NaH_2PO_4. According to the researches of Lehmann, however, there can be no doubt, that in many cases, free hippuric and lactic acids exist in the urine, and consequently assist in giving it its acid reaction.

The urine is alkaline during digestion, owing to the increased elimination of alkaline phosphate, such as Na_3PO_4 derived from the food.

3. *Fermentations of urine.*—Two fermentations occur : 1st, the *acid ;* 2d, the *alkaline.*

When the urine has been left at rest, especially under the influence of a moderate degree of heat, its acid reaction becomes stronger; and distinct crystals of uric acid are often deposited on the sides and bottom of the glass. This increase of its acidity usually goes on for some days, and may even continue in rare instances for two or three weeks. The acidity, however, at last begins suddenly to diminish, and gradually disappears. The urine now becomes lighter in colour; a whitish, iridescent pellicle forms on its surface; and the presence of ammoniacal odour indicates that it has become alkaline. A deposit is thrown down consisting of the ammoniaco-magnesian or triple phosphate, phosphate of lime, and urate of ammonia. This change, the alkaline fermentation, is owing to the decomposition of the urea into carbonate of ammonia. Urea unites with the element of water thus :—

$$\underset{\text{Urea.}}{CN_2H_4O} + 2\,\underset{\text{Water.}}{(H_2O)} = \underset{\text{Carbonate of ammonia.}}{(NH_4)_2CO_3}$$

The urine is thus rendered alkaline, and the earthy phosphates are precipitated,—the phosphate of lime as such, and the phosphate of magnesia as the triple phosphate of ammonia and magnesia ($MgNH_4PO_4 + 6\,H_2O$).

4. *Quantity in twenty-four hours.*—The determination of the quantity of urine passed in a given time forms the basis of all quantitative investigations, and must therefore not be overlooked. In all analyses of the urine, the quantity of the fluid, and the time during which it is collected, must be taken into consideration. The time usually adopted is twenty-four hours. The quantity can be determined either by weight or measure; but measure is almost invariably employed for the purpose. The cubic centimetre we take as a standard of unity; one thousand cubic centimetres are equal to a litre, and one litre of distilled water weighs a thousand grammes. When we have learnt the specific gravity of urine, the quantity of which has been ascertained, we may readily arrive at a knowledge of its weight, by simply multiplying the number of ascertained cubic centimetres by the specific gravity of the urine. Thus 1000 C. C. of urine of 1·030 sp. gr. will weigh 1030 grammes. The urine is measured by means of graduated glass jars of different sizes. Care should also be taken in all examinations of urine that the glass vessels are kept quite clean, because a small amount of decomposing

organic matter is sufficient to induce the alkaline fermentation in a few hours.

5. *Amount of solid matter.*—For this purpose we require an accurate chemical balance, several small porcelain crucibles or capsules, a water bath formed of copper plate, a hot air bath or chamber, and a shallow vessel containing a little strong sulphuric acid which can be placed under a bell-jar. The method of estimating the amount of solid matter is as follows :—

(1.) Measure ten cubic centimetres * in a carefully weighed porcelain capsule.

(2.) Evaporate to dryness at a low temperature over a water bath.

(3.) Dry as thoroughly as possible, and afterwards place the capsule in the hot air chamber for several hours.

(4.) Remove the capsule from the chamber, and place it over the sulphuric acid under the bell-jar for two hours.

(5.) At the end of that time remove it, and weigh rapidly. The difference in weight from that of the urine used, gives the amount of solid matter in the urine.

The object of this process is to remove the water as thoroughly as possible. An example will illustrate the calculation :—

Weight of capsule alone	-	30·62 grammes.
Weight of capsule with residue		30·84 „
Weight of residue	-	0·22 „

Therefore in 10 C. C. of the urine examined there are 0·22 grammes of solid matter. It is important, if a very accurate estimate is required, to weigh several times after the residue has been placed over the sulphuric acid, and not to finish the process until there is almost no difference (say one or two millegrammes) between the two weights. †

6. *Amount of organic and inorganic matter.*—That the residue obtained by evaporating a certain quantity of urine consists partly of organic matter may be easily shewn by the fact that it chars on being strongly heated. If a white heat be applied for some time, the blackened appearance is removed, and a

* The symbol for cubic centimetres is C. C.; for grammes, grms.; for millegrammes, m.grms., &c.

† A still more accurate method is described by Neubauer and Vogel in their "Guide to the Qualitative and Quantitative Analysis of the Urine," New Sydenham Society. 1863. P. 153. The method of examining urine described in the text is chiefly that of Neubauer and Vogel.

white ash is left behind. This white ash consists of inorganic salts. The mode of determining the amount of organic and inorganic matter is as follows :—

(1.) Evaporate 10 C. C. of urine in a porcelain capsule.

(2.) When the residue is dry, scrape it out with a small platinum knife, and place it in a weighed platinum capsule (along with from 10 to 13 drops of strong nitric acid or with a known weight of spongy platinum), in which it is to be heated, at first gently, but afterwards strongly. We thus obtain a white ash, free from carbon. The addition of nitric acid converts the urea into nitrate of urea, which when heated is first decomposed into carbonic acid and nitrate of ammonia, and finally escapes as water and nitrous oxide gas. By using nitric acid as an oxidising agent we save time, for the urea, which forms the greatest part of the residue, and produces much carbon at the ordinary red-heat, is thereby removed, and a portion of the remaining carbon more readily oxidises and burns off under the action of the nitrate of ammonia which is formed. We must, however, carefully avoid adding too much nitric acid, and using too great a heat, so as to avoid losing chlorine and phosphorus.

(3.) Weigh the ash with the crucible, and subtract from it the weight of the crucible alone, and the difference gives the actual amount of incombustible salts. Example :—

Crucible with ash - - -	24·656	grammes.
Crucible alone - - - -	24·524	„
Weight of ash in 10 C. C. of urine	·132	„

7. *Amount of water.*—The loss of weight, after evaporating, at a low temperature, 10 C. C. of urine represents the amount of water in 10 C. C.

8. *Specific gravity.*—This may be determined in three ways :—

(1.) *By the specific gravity bottle.*—Ascertain the weight of the bottle alone, then the weight of the bottle filled with distilled water, and thirdly, the weight of the bottle filled with urine. In each operation the bottle must be quite full and carefully wiped dry. Then subtract from each of the last two the weight of the bottle. The proportion then is : weight of water : weight of urine : : specific gravity of water : specific gravity of urine. Example :—

Weight of bottle = 12 Grammes.
Weight of bottle + water = 45 „
Weight of bottle + urine = 46 „

Therefore 33	:	34	::	1000	:	1030.
Weight of water.		Weight of urine.		Specific gravity of water.		Specific gravity of urine.

The chief objection to this process is that it is tedious, but with care great accuracy may be attained.

(2.) *By the urinometer.*—We obtain only an approximative knowledge of the true specific gravity of the urine by the aid of the urinometer. This instrument consists of a glass float bearing a graduated stem, and kept upright by a little ball at the bottom containing mercury. It should be graduated so that the zero of the scale be on a level with the surface of the fluid when placed in distilled water, and the degrees should go as high as 1050.

To determine the specific gravity of the urine by means of the urinometer, a proper cylindrical glass is filled with the urine, all froth removed by means of a glass-rod, and the clean urinometer allowed to sink gently into the fluid. The glass should be wide enough to allow the instrument to float freely in the urine, and not to touch its sides. Bring the eye on a level with the surface of the fluid, and read off the scale at the lower level of the curve ⌣ formed by capillary attraction ; always read off at that level.

(3.) By glass beads of such a weight that they will float exactly at the surface in fluids of various specific gravities. These beads are numbered from 5 to 50, with many intermediate numbers. If bead No. 20 floats in the fluid so that its upper surface is exactly on a level with the surface of the fluid, the specific gravity of the fluid is 1020.

9. *Christison's method of ascertaining the amount of solid matter from the specific gravity.**—The rule is to multiply the last two figures of the specific gravity, ascertained as above, by 2·33, (a number ascertained from numerous experiments) and the quotient gives the amount of solid matter in 1000 parts of urine. *Example:*—A man passes 46 ounces of urine in 24 hours. Specific gravity, 1025. How much solid matter is excreted?

* Sir Robert Christison, Bart., Tweedie's Library of Medicine, vol. iv. p. 248, line 6.

—25 × 2·33 = 58·25 oz. of solid matter in 1000 oz. Then 1000 : 58·25 : : 46 : 2·6795 oz. *Answer.*—Amount of solid matter excreted by kidneys in 24 hours, 2·6795 oz.

Volumetric Analysis.

It is extremely convenient to estimate the amount of certain constituents of the urine by volumetric processes because the analysis is simplified and time is saved. It is therefore essential first, that we understand the theory of the process, for which we are indebted to Gay-Lussac.

Theory of the process.—It consists in submitting the substance to be estimated to certain well-known reactions, using for such reactions, solutions of known strength, and from the quantity of solution employed, calculating the weight of the substance to be estimated according to the laws of equivalence. For example :—

Suppose that it is desirable to know the quantity of pure silver contained in a shilling. The coin is first dissolved in nitric acid, forming a bluish solution, containing silver, copper, and probably other metals. It is known that chlorine combines with silver in the presence of other metals to form chloride of silver, AgCl, which is insoluble in nitric acid. The proportions in which the combination takes place are 35·5 of chlorine to every 108 of silver ; consequently, if a standard solution of pure chloride of sodium is prepared by dissolving 58·5 grammes of the salt (*i.e.*, 1 eq. sodium = 23, 1 eq. chlorine = 35·5 = 1 eq. chloride of sodium, 58·5) in so much distilled water as will make up exactly 1000 C. C. by measure ; every C. C. of this solution will contain exactly enough chlorine to combine with 0·108 grammes of pure silver to form chloride of silver, which precipitates to the bottom of the vessel in which the mixture is made. In the process of adding the NaCl solution to the silver, drop by drop, a point is at last reached when the precipitate of AgCl ceases to form. Here the process must stop. On looking carefully at the graduated vessel from which the standard solution has been dropped, the operator sees at once the number of C. C. which have been necessary to produce the complete precipitation of the silver, and as each C. C. equals ·108 grammes of silver, it is easy to calculate the quantity of the latter present in the shilling.

We therefore require in every volumetric process :

1. A solution of the re-agent, the chemical equivalence of which is accurately known. This we term the standard solution (symbol S.S.).

2. A graduated vessel from which portions of it may be accurately delivered—the burette.

3. The decomposition which the solution produces with any given substance is usually of such a character that its termination is unmistakable to the eye, and thereby the quantity of the substance with which it has combined accurately determined. Occasionally, however, we use another solution which produces a characteristic reaction with the standard solution, and which thus informs us when we have added excess of the standard solution. This is termed the *indicator*.

Apparatus required. 1. *The graduated pipette.*—It is made of glass. It serves for measuring the fluid which is to be investigated; and when filled to the neck, where it is marked by a single stroke or mark, contains 50, 20, 15, 10, 4, or 3 C.C. In using it, its point is introduced into the fluid, and suction made until the fluid has risen above the level of the mark in the neck; the upper opening is then closed with a moist finger, the pipette dried outside to remove any adherent fluid, and the finger slightly raised to admit a little air, and to allow the fluid to escape until it reaches the level of the mark, the surface of the fluid being kept on the same level as the eye. When the fluid has fallen to this point, the pipette is again firmly closed with the finger, and its contents may now be allowed to run out into any convenient vessel, such as a beaker.

2. *Flasks and Jars.*—These may be graduated from $\frac{1}{10}$th litre to 5 litres. There is usually a mark across the neck indicating the volumetric capacity. It is convenient to have these flasks so arranged that the volumes shall be whole numbers—not $1\frac{2}{3}$ litres, $2\frac{3}{4}$ litres, but 1, 2, 3 litres, and so on.

3. *Mohr's Burette.*—This instrument (Plate XXI. fig. 26, *a*, *c*) consists of a glass tube provided below with a caoutchouc tube *a*, which is closed by a spring clamp *e*. Two or more of these may be fixed into a wooden or iron frame *g*, *h*, *m*, *m*, so as to hang down perpendicularly.

In using it, the pipette is filled up to zero with the volumetrical fluid, the urine to be tested measured out into the beaker glass *l*, and the volumetrical solution then allowed to run out into the glass beaker *l*, by pressing on the clip, and towards the end of

the experiment to drop into it, until the proper quantity has been added.* By this arrangement we secure both a rapid flow of the fluid, and also its flow in single drops. In investigations which require some time for their completion, two or more of these burettes are employed ; when completely or half-filled, they are fixed in the stand and there left, their upper opening being closed with a cork to prevent evaporation. Certain standard solutions act injuriously on the caoutchouc and clamp (such as nitrate of mercury in the urea process), and destroy them. In these instances it is advisable to use a burette, having a glass stop cock.

Mode of conducting the process.—In carrying out the volumetrical method of analysis, first carefully prepare the solutions required for the purpose, for upon these solutions the correctness of the analyses depends. Special directions for this object will be given under the head of each particular process. The solutions must always be prepared and used at a given temperature, as their volume varies considerably under the action of heat. Care is also required in reading off the level of the fluid in the different kinds of measures used. Bubbles of air must be removed by a glass rod, so that the surface of the fluid be perfectly level. This point is obtained in the case of the pipette by allowing it to hang freely. We must also allow for the capillarity of the tube. When we examine the curve, especially by transmitted light, several zones are readily distinguished in it. The measurements are most accurate, when (the pipette or the burette having been placed in a perpendicular condition) the eye is brought to a level with the under border of the lowest zone, and the graduation of the tube corresponding with it then read off. This border is most distinctly marked and seen by transmitted light.

When the urine to be tested has been measured, and the pipette or the burette filled with the volumetrical solution, we first of all allow the solution to run slowly out, and at last to pass drop by drop into the urine, until the operation is completed. When the point of completion is shewn in all parts of the fluid, by some distinct reaction, or by the use of an *indicator*, we are sure that the process followed is good ; but if this be not the

* The volumetrical fluid is usually placed in the burette and the urine in a beaker or porcelain capsule, but occasionally, as in the diabetic sugar process, the reverse is the case.

case, then we must test the mixture again and again towards the conclusion of the experiment, until the right point has been attained.*

Detection and Estimation of the Individual Inorganic Constituents of Healthy Urine.

Under this head we shall include chlorides, sulphates, phosphates, iron, ammonia, and silicic acid.

1. *Chlorides* (p. 28).—Nitrate of silver always serves as a test for the presence of chlorides in the urine, giving a white curdy precipitate. The phosphoric acid in the urine also throws down a precipitate with nitrate of silver; but this precipitate—phosphate of silver—is soluble in nitric acid, which the chloride of silver is not. Consequently, in testing the urine for chlorine we must, before the nitrate of silver is dropped into it, render the mixture strongly acid by the addition of one or two drops of nitric acid. This will prevent the phosphate of silver being thrown down.

The chlorine exists chiefly in combination with sodium, but a small amount is in the compound of chloride of potassium. The *soda* may be demonstrated in the urine by giving a yellow colour to the inner blow pipe flame, and the *potash* by giving a yellow precipitate of octahedral crystals of the double chloride of potassium and platinum on the addition of the tetrachloride of platinum to an acid and alcoholic solution of the ash.

The amount of chlorides varies considerably. They are much diminished in all acute febrile diseases, the quantity sinking to a minimum, so as sometimes to form scarcely one hundredth of its normal standard. The quantity increases as the disease passes away, and during convalescence is occasionally greater than normal. The presence of a large quantity of chlorine, 6-10 grammes daily, indicates good digestion; a small quantity, under five grammes, weak digestion, provided always that the diet of the patient is not such that only very little chlorine is ingested.

2. *Sulphates* (p. 29).—The sulphates yield, with chloride of

* In this work, we shall give the volumetric processes for only a few of the constituents of urine, namely, those most important physiologically or pathologically. For details as to the others, reference is made to Neubauer and Vogel, and other works.

barium or nitrate of baryta, a precipitate which is insoluble in mineral acids, and easily detected even when the solution is exceedingly diluted. Consequently, in testing the urine for its sulphates, we first of all render it strongly acid by the addition of a drop of nitric acid or hydrochloric acid, for the same reasons as given in the case of chlorides, and then add to it a solution of chloride of barium or nitrate of baryta. A heavy precipitate falls of sulphate of baryta. If, therefore, we take a certain volume of urine, say 10 C. C., and add to it an equal or sufficient quantity of chloride of barium and hydrochloric acid, we obtain, from the greater or less quantity of precipitate which is thereby thrown down, an approximative estimate of the amount of sulphates present in it.

3. *Phosphates* (p. 28). — These consist of phosphates of the alkalies, and phosphates of the alkaline earths. The latter are insoluble in an alkaline fluid, and consequently are always precipitated when the urine becomes alkaline (p. 262).

Tests for the phosphates.—(1.) Chloride of barium, or nitrate of baryta, give a precipitate of phosphate of baryta, soluble in mineral acids.

(2.) Ammonia, or caustic potash, or caustic soda, give a precipitate of phosphates.

(3.) Perchloride of iron throws down from a solution of phosphates containing free acetic acid, a yellowish-white precipitate of perphosphate of iron.

(4.) Acetate of uranium added to urine containing a few drops of free acetic acid, gives a light yellow or lemon-coloured precipitate, consisting of uranium and ammonium double phosphate.

(5.) Molybdate of ammonia, along with a few drops of nitric acid on boiling yields a brownish, greenish, or canary-yellow precipitate of the phospho-molybdate of ammonia. This is an exceedingly delicate reaction.

Volumetric Process for Phosphoric Acid.

It is often important to determine with accuracy the amount of phosphoric acid excreted in a certain period of time. This is best accomplished by the volumetric process which depends on the fact that a precipitate of uranium and ammonium double phosphate ($2Ur_2O_3NH_4PO_4$) is immediately formed, when a hot solution of a phosphatic salt which is soluble in water or acetic

acid, is treated with a solution of acetate or nitrate of uranic oxide in presence of free acetic acid.* The phosphate of uranic oxide thus thrown down, appears as a whitish-yellow, passing even into a greenish, precipitate; it is completely insoluble in water and acetic acid, but soluble in mineral acids. The exact point of the completion of the reaction cannot be ascertained in the fluid, on account of the slimy character of the precipitate, and of the slowness of its deposition; consequently, in order to determine whether or not the whole of the phosphoric acid is precipitated, a small excess of uranic oxide must be added,—the presence of this salt being readily shewn by the highly sensitive reaction of the salts of uranic oxide with ferrocyanide of potassium, which gives a reddish-brown precipitate. The ferrocyanide of potassium thus serves as an indicator.

It is necessary, in the first place, to prepare with great care the standard solutions.

(*a.*) *Standard phosphoric acid-solution.*—This solution should be so constituted as to resemble the urine as nearly as possible, as regards the amount of phosphoric acid; 50 C. C. of it should contain 0·1 gramme of phosphoric acid. It may be readily prepared from chemically pure phosphate of soda, which has not undergone efflorescence. The pure crystals are rubbed down as fine as possible, dried between folds of bibulous paper, 10·085 grammes weighed and dissolved in a litre of water. 50 C. C. of this solution contain exactly 0·1 gramme of phosphoric acid.

(*b.*) *Acetate of soda-solution.*—It has been found 0·5 gramme of acetate of soda is, under all circumstances, sufficient for 50 C. C. of urine. Consequently, 100 grammes of acetate of soda are dissolved in 900 C. C. of water, and the solution brought up to a litre by the addition of 100 C. C. of concentrated acetic acid. In the volumetrical process, 50 C. C. of urine are treated with 5 C. C. of this acid solution of acetate of soda.

(*c.*) *Solution of uranic oxide.*—Pure commercial uranic oxide, is dissolved in pure acetic acid, free from all empyreumatic matters, the solution diluted, and its strength tested with the standard phosphate of soda-solution (*a*). One C. C. of it should precipitate, and indicate the presence of, only 0·005 gramme of phosphoric acid. 50 C. C. of the phosphoric acid-solution (*a*) = 0·1 gramme of phosphoric acid, would consequently require exactly 20 C. C. of the uranic oxide solution; this

* Neubauer and Vogel, pp. 191-193.

solution, therefore, must, in the first place, contain 0·4023 gramme of uranic oxide for the precipitation of the phosphoric acid, and, secondly, a slight excess of uranic oxide for the indication of the completion of the reaction.

50 C. C. of the solution of phosphoric acid require 20 C. C. of the uranic oxide-solution, which again must indicate and precipitate 5 milligrammes of phosphoric acid. If, for example, we employ 18·0 C. C. of the uranic oxide-solution to 50 C. C. of phosphoric acid-solution, we must add to each 180 C. C. of the same 20 C. C. of water. For this purpose we measure off 1 litre of the uranic oxide-solution, and add to it the quantity of water required. In the case supposed, 111·2 C. C. of water must be added to 1000 C. C. of uranic oxide-solution to produce the required degree of strength.

Thus, if we have a second time used 19·8 C. C. of uranic oxide-solution to 50 C. C. of phosphoric acid solution (0·1 gramme of phosphoric acid), we add to each 198 C. C. of the same 2 C. C. of water, and make a new and final test with the phosphate of soda-solution.

The uranic oxide-solution, each cubic centimetre of which precipitates 5 milligrammes of phosphoric acid, and which also contains a small excess of uranic oxide for the final re-action, must contain 20·3 grammes of pure uranic oxide in a litre.

Process for the whole of the phosphoric acid with acetate of uranium, ferrocyanide of potassium being used as an indicator.

1 C. C. of S. S. = 0·005 grammes of phosphoric acid.

(1.) Place 50 C. C. of filtered urine in a beaker.

(2.) Add to it 5 C. C. of a solution of sodium acetate.

(3.) Drop in standard solution of uranium acetate, until a drop gives a faint brown colour when mixed with a drop of potassium ferrocyanide, on a porcelain plate.

(4.) Boil and test again. If necessary, add a few drops more of the S. S. until the brown colour again appears immediately on testing.

Example.—Patient passes in 24 hours 1000 C.C. of urine ; 25 C. C. of S. S. are used in volumetric process for phosophoric acid. How much phosphoric acid is excreted :—·005 × 25 = ·125 grammes in 50 oz. Then 50 : ·125 : : 1000 : 2·5 grammes, the quantity in 1000 C. C. of urine.

Process for estimating the amount of phosphoric acid united with the alkaline earths.

(1.) Take 100 C. C. of filtered urine, and make it alkaline with ammonia. The earthy phosphates are thus precipitated.

(2.) Let the urine stand for 12 hours.

(3.) Collect the earthy phosphates on a filter, and wash with ammonia water.

(4.) Wash precipitate into a beaker, heat and dissolve in a few drops of acetic acid.

(5.) Add 5 C. C. of acetate of sodium solution, and add water to make up volume to 50 C. C.

(6.) Proceed with acetate of uranium solution as before, and make the necessary calculation.

Taking the previous example, we find that :—

The whole of the phosphoric acid, as determined by acetate of uranium process, is	2·5 grammes.
Phosphoric acid with the earths required 5 C. C. of S. S. Therefore, ·005 × 5 = ·025 grammes in 100 C. C. of urine. Patient passed 1000 C. C. Therefore, in 1000 C. C. we find of phosphoric acid united to the alkaline earths, -	0·25
Phosphoric acid with alkalies, -	2·25
	2·5 grammes.

4. *Iron.*—This is rarely found in urine, and only in very minute quantities. It probably exists in the colouring matter. For testing and ascertaining the presence of iron in the urine, the ash obtained from the urine is always employed. Dissolve in a few drops of hydrochloric acid. Boil with a drop of nitric acid, and add a drop of sulphocyanide of potassium, thereupon the fluid will assume a reddish colour, and if a considerable quantity of iron be present, a deep dark-red colour. When mere traces are present, the change of colour is best observed by placing the tube over a white ground.

5. *Ammonia.*—According to Neubauer and Vogel, a small amount of free ammonia is present even in acid urine, but its quantity is so small as to render its detection extremely difficult. It is of no importance.

6. *Silicic acid.*—This acid has been detected in very small amount by incinerating the ash of urine with sodium and

potassium carbonate, dissolving the ash in water, and acidulating with hydrochloric acid. On again evaporating to dryness, the silicic acid remains behind in a pure state.

Detection and Estimation of the Individual Organic Constituents of Healthy Urine.

These are urea, uric acid, hippuric acid, creatin and creatinin, xanthin, and benzoic, phenylic, damaluric, damolic, and succinic acids.

1. *Urea* (p. 14 and p. 25).—This substance may be prepared from the urine by first precipitating all the phosphates by means of baryta, filtering, evaporating the filtrate, and treating the residue with alcohol. This alcoholic solution is evaporated to dryness, and the product again treated with pure alcohol. We thus obtain an alcoholic solution of urea, which crystallises out on evaporation. The most important salt of urea is the nitrate, which may be obtained by mixing a concentrated solution of urine or urea with concentrated and pure nitric acid. It then appears as white plates.

Tests for Urea.—(*a.*) When the quantity of urea is small, the formation of nitrate of urea may be observed under the microscope, and in the following way:—One end of a little bit of thread is laid in the drop which is to be tested for urea; the drop itself and one-half of the thread is then covered with the glass, and the other end of the thread moistened with a drop of pure nitric acid. In this way, the two fluids being gradually mixed together, we may see the formation of crystals of rhombic plates or short prisms, as well as numerous complicated forms.

(*b.*) When a solution of nitrate of mercury is added to urine, we obtain a white floculent precipitate, which varies in composition according to circumstances. It may, according to the quantity of urea present, consist of mercuric oxide and nitrate of urea, or urea combined with mercuric nitrate and mercuric oxide. Upon this reaction, however, the volumetric process is based.

Volumetric Process for Urea.

When a dilute solution of urea is added to an equally dilute solution of nitrate of mercury, and the free acid neutralised by carbonate of soda, a white precipitate is obtained. After all the urea has been precipitated, we reach a point where the yellow-coloured hydrated oxide of mercury is thrown

down by the carbonate of soda, carbonic acid escaping with effervescence. It has been found by analysis that the urea is thrown down in combination with oxide of mercury, and that the precipitate contains four equivalents of oxide of mercury to one equivalent of urea. The exact point, or rather the exact point has just been over-stepped, when all the urea is precipitated, and is known by the formation of a yellow pellicle on the surface of a drop of carbonate of soda mixed with a drop of the fluid being examined for urea.

Preparations of standard solutions. (*a.*) *Standard solution of urea.*—Four grammes of pure urea, dried at 100° C., are dissolved in water, and diluted until the volume of the fluid equals 200° C. C. Thus 10 C. C.=·2 grammes of urea.

(*b.*) *Standard solution of nitrate of mercury.*—Pure oxide of mercury best serves for the preparation of the mercury solution. Commercial oxide of mercury may be obtained sufficiently pure for the purpose. An oxide of mercury which leaves no visible residue when heated on platinum foil is fitted for the purpose. Of this oxide 77·2 grammes, dried at 100° C. are taken by weight, dissolved under a gentle heat with the smallest possible quantity of nitric acid in a porcelain basin, evaporated to a syrup, and then diluted with water up to a litre. Should any basic salt separate, a few drops of nitric acid are dropped into it until the precipitate re-dissolves.

The next step is to graduate the prepared solution of mercury by means of the standard solution of urea. For this purpose 10 C. C. of the urea-solution are measured off and placed in a beaker, the dilute mercury solution is then added to it, until a few drops of the mixture, added to a drop of carbonate of soda on a watch-glass, give a yellow colour. If, for example, to obtain this reaction, we use 19·25 C. C. of the mercury solution, we add to each 192·5 C. C. of the mercury solution, 7·5 C. C. of water, and thus get 200 C. C. of a solution, 20 C. C. of which will precipitate the urea from exactly 10 C. C. of urea solution (that is ·2 grms.). Thus 10 C. C. of mercuric nitrate solution will correspond to ·1 grms. of urea.

Process for estimating Urea with Nitrate of Mercury, Carbonate of Soda being used as an Indicator.

(1.) If albumin be present in the urine, separate it by boiling and filtration.

(2.) Mix the urine with half its volume of a solution called "baryta mixture" (composed of two volumes of solution of Barium hydrate with one volume of Barium nitrate, both saturated in the cold).

(3.) Filter to get rid of Barium sulphate and phosphate.

(4.) Take 15 C. C. of filtrate (=10 C. C. of urine) and place in a beaker.

(5.) Drop in S. S. till precipitate ceases, testing the mixture from time to time with a solution of sodium carbonate, until a faint yellow reaction is obtained.

We thus obtain a knowledge of the quantity of urea in 10 C. C. of urine.

Example.—Patient passes in 24 hours 1000 C. C. of urine, 14 C. C. of solution of mercuric nitrate are employed. How much urea is excreted? 1 C. C. of mercuric nitrate solution =·01 grms. of urea ∴ 14 C. C. = ·14 grms. in 10 C. C. of urine. Then 10 : ·14 : : 1000 : 14 grms. of urea, the quantity in 1000 C. C. of urine.

*Corrections for urines containing more or less than two per cent. of urea.**—The reaction between mercuric nitrate and carbonate of soda is exact only for fluids containing two per cent. of urea, and we require 30 C. C. of S. S. for complete precipitation of the urea in every 15 C. C. of urine, as well as for the reaction with sodium carbonate. When the urine contains more than two per cent. of urea the reaction takes place too soon, when it contains less the reaction is delayed.

(*a.*) *With more than two per cent. or excess of urea.*—When double the volume of mercurial solution has been used, and no reaction set in, 1 C. C. of distilled water is added to the mixture for every additional 2 C. C. of the mercurial solution used, and thus the proportion of urea is maintained at two per cent. Thus, if 30 C. C. of solution of mercuric nitrate are added to 15 C. C. of urine, and the reaction is not seen, 1 C. C. of distilled water is added, and the process is continued. If the reaction set in when 10 C. C. more, or 40 C. C. in all, of the mercurial solution have been used, the 5 C. C. of distilled water added (*i.e.* 1 C. C. of water for every 2 C. C. of the excess over 30 C. C.) will, with the original 15 C. C. of urine, make 20 C. C., and the mercurial

* Corrections must also be made, if great accuracy be desired, for the chloride of sodium, and carbonate of ammonia, but as these are not very important, reference is made to Neubauer and Vogel, p. 185.

solution will have been employed on a urine containing two per cent. of urea.

(*b.*) *With less than two per cent of urea.*—If the urine contain less than two per cent. of urea, subtract ·1 C. C. from every 5 C. C. of mercurial solution less than the normal 30 C. C. Thus, if with 15 C. C. of urine, the reaction with sodium carbonate is got on using 20 C. C. of solution of mercuric nitrate, ·2 C. C.—that is ·1 × 2, are deducted, and 19·8 C. C. taken as correct.

2. *Uric acid* (pp. 13, 357).—The presence of this acid may be readily demonstrated in urine by placing a few ounces in a conical glass, adding a few drops of hydrochloric acid, and allowing it to stand for forty-eight hours. Uric acid then crystallises out, and appears on the surface of the fluid and adhering to the bottom and sides of the glass. When examined microscopically, the crystals will be found as represented in Plate I. figs. 1 and 2.

The murexide test for uric acid.—Place a few drops of urine on a large flat porcelain lid. Add a drop of nitric acid. Evaporate nearly to dryness, and then bring a glass rod dipped in a solution of ammonia over the residue. A splendid purple-red or violet colour of murexide is produced. In this test the nitric acid frees uric acid from its union with bases, and converts it into two substances termed alloxan and alloxantin (p. 13). A compound called murexide or purpurate of ammonia is then formed by the union of the ammonia with these two uric acid derivatives.

*Schiff's test for uric acid.**—Dissolve the suspected powder in sodium carbonate, and place a drop of the solution on a bit of blotting paper moistened with nitrate of silver solution ; if uric acid be present, a brown spot appears, carbonate of silver being reduced to oxide by uric acid at ordinary temperatures.

Garrod's test for uric acid present in small quantity.†—This test is more especially applicable to the detection of uric acid in the blood. Take about two drachms of the serum and place it in a flat glass dish or watch-glass. To this add twelve drops of ordinary strong acetic acid, which will cause the evolution of a few bubbles of gas. When the fluids are mixed, introduce two or three threads of unwashed cotton. Allow the glass to stand on the mantel-piece, or on a shelf in a warm room, for from

* Schiff. Ann. Ch. Pharm. cix. 65.

† Garrod On Gout. London. 1863.

thirty-six to sixty hours, until its contents set, from evaporation. If the cotton fibres be then removed and examined microscopically with a half-inch object-glass, they will be found covered with crystals of uric acid, if this agent be present in the serum. The crystals form on the thread somewhat like masses of sugar-candy on string.

3. *Hippuric acid* (p. 15).—This acid exists in very minute quantity in human urine, more especially after a person has taken benzoic acid, toluene, cinnamic, or mandelic acids, but it may be readily prepared by treating the urine of a cow or horse with excess of lime water, and thus precipitating it as hippurate of lime. Evaporate to 1-10th of the original volume of the urine, and add hydrochloric acid. Hippuric acid crystallises out impure, but the crystals may be obtained colourless and semi-transparent by dissolving them in water in the presence of animal charcoal, and allowing them again to crystallise out (Plate I. fig. 5).

4. *Creatin and creatinin* (p. 18).—Urine contains only a small quantity of creatin and creatinin, so that a large amount of the fluid is required for their demonstration. The mode of separating these from urine is as follows:—Three hundred C. C. of fresh urine are neutralised with milk of lime, and the phosphoric acid then thrown down by a solution of chloride of calcium. Filter and quickly evaporate to dryness in a water-bath. The residue thus obtained is extracted with absolute alcohol, allowed to stand for some hours, and again filtered; the clear fluid is then treated with a few drops of a concentrated solution of chloride of zinc free from acid. The mixture becomes turbid, and the separation of the creatinin-chloride of zinc is completely effected in forty-eight hours. The compound is washed on a filter with spirits of wine, dried, and microscopically examined (Plate I. figs. 11 and 12).

To obtain the creatinin in a pure state, dissolve the zinc compound in a small quantity of boiling water, and separate the oxide of zinc and hydrochloric acid by boiling the fluid with freshly-precipitated and well-washed hydrated oxide of lead. The filtered liquid is rendered colourless by boiling with animal charcoal, and evaporated to dryness. The residue, which consists of a mixture of creatinin and creatin, is then treated with cold strong spirits of wine, whereby the creatinin is dissolved and the creatin left.

Should the urine operated upon contain albumin, the albumin must be previously separated from it by coagulation.

5. *Xanthin* (p. 16).*—The mode of preparing this substance from urine is as follows:—Fresh, healthy urine, in quantity not less than from 100 to 200 pounds, is evaporated in a water-bath to from one-sixth to one-eighth of its original volume, and its phosphoric acid removed by precipitation with baryta-water. The filtrate is again evaporated until the salts are crystallised out of it; the mother-liquor thus obtained is then well diluted with water, a solution of acetate of copper added, and boiled for some time. A dirty-brownish precipitate is thus obtained, which is first decanted and then washed on the filter with cold water until all chlorine-reaction has disappeared. By treating this precipitate with hot nitric acid, we obtain a brownish solution, from which the impure xanthin-silver compound is precipitated by nitrate of silver. The crystalline compound, after being washed, is dissolved in boiling dilute nitric acid; any remaining flocculi of chloride of silver are removed by filtration, and the filtrate set aside and allowed to crystallise slowly. The collected crystalline silver-compound is freed from nitric acid by digestion with an ammoniacal solution of silver; the washed precipitate diffused through water, boiled, and the compound decomposed by sulphuretted hydrogen. The boiling filtered solution deposits, when concentrated, coloured flocculi of xanthin, and the remainder is obtained by further evaporation. The preparation thus obtained is, however, always much discoloured; but by solution in strong hydrochloric acid and treatment with animal charcoal, the purification is readily effected. The filtrate, thus freed from colour, yields, when evaporated, hydrochlorate of xanthin, from which pure xanthin may be obtained by repeated treatment with ammonia, and by subsequent removal of the chloride of ammonium by washing with cold water.

6. *Benzoic, phenylic, taurylic, damaluric, damolic, succinic, oxalic, formic, lactic, and acetic acids.*—Human urine contains only a very small and variable quantity of these acids. Acetic and butyric acids are usually to be found in decomposing urine. It is not within the scope of this work to describe the various processes by which these substances can be obtained from urine, and reference is made to larger works on Physiological Chemistry.

* Neubauer and Vogel, p. 24.

Detection and Estimation of the Abnormal Constituents of Urine.

We shall consider the following: albumin, sugar, bile pigment, bile acids, fat, kiestein, lactic, acetic, and butyric acids, sulphuretted hydrogen, allantoin, leucin, and tyrosin.

1. *Albumin.*—This substance is occasionally present for a short time in healthy urine, but as a rule its presence is indicative of disease of the kidneys. It is therefore of the greatest importance to be able to detect it even in minute quantity. Albumin is always present in urine containing blood.

Tests for albumin. (*a.*) *Heat.*—In the first place, test the reaction of the urine with litmus paper. If it be alkaline, or neutral, add to it a few drops of acetic or nitric acids; if very acid, carefully neutralise with a little dilute ammonia. Boil a small quantity in a test tube. If albumin be present in small amount, the fluid will become turbid when the heat exceeds 68° C.; if it be abundant, there will be a distinct coagulation. It is important to remember that if the urine be either alkaline or neutral, coagulation may not take place—the albumin, if present in small quantity, uniting with the alkali. On the other hand, if a small amount of albumin be present in a large quantity of water, and there be excess of acid, no coagulation may follow, because a combination of albumin with the acid may be formed, which is soluble in water. Another fallacy to be remembered is, that upon boiling certain varieties of urine, a precipitate of earthy phosphates takes place, which, however, can be readily distinguished by the addition of a little dilute nitric acid, which dissolves the phosphates, but not the albumin.

(*b.*) *Nitric acid test.*—On adding nitric acid to urine a white turbidity occurs if albumin be present in small, and distinct coagulation if present in large, amount. Sometimes, however, no coagulation is obtained because nitrate of albumin is formed, which is soluble in a large quantity of water; in other specimens of urine, a precipitate of nitrate of urea may be formed, which, however, is readily detected by means of the microscope; while in the urine of patients in the habit of taking copaiva, cubebs, and other oleo—and resinous—medicines, a white turbidity appears, which, however, does not sink to the bottom of the test tube as albumin does, but will remain for many hours suspended in the fluid.

(*c.*) *Ferrocyanide of potassium test.*—When to a well-filtered urine, acidulated with acetic acid, a weak solution of ferrocyanide of potassium (5 grains to the ʒi) is added, there is a white precipitate. If there be a large quantity of mucus in the urine, this test is not serviceable.

In testing for albumin, therefore, it is better to employ both heat and nitric acid than either alone, and if the above sources of fallacy are borne in mind, there is usually no difficulty in detecting even minute traces of albumin.

Estimation of the quantity of albumin by weight.—Place 20 C. C. of urine, diluted with 80 C. C. of water, in a beaker, and allow the albumin to coagulate in a water bath. Collect the coagulum on a filter, wash and dry it at 100° C., weighing occasionally until there is no appreciable difference between two weighings. This is albumin with inorganic matter. Incinerate, and collect and weigh the ash. Deduct this from the weight of albumin + inorganic matter, and the difference will be a very near approximation to the amount of albumin. *Example.*—Patient passes 1000 C. C. of urine in 24 hours. 20 C. C. yielded ·454 grammes of albumin + ash. After incineration, the ash was found to weigh ·0015 grammes ∴ ·454—·0015=·4525 grammes in 20 C. C. In 1000 C. C. therefore, the amount of albumin would be ·4525 × 50 = 22·625 grammes.

There is also a volumetric process for albumin depending on the fact that albumin is precipitated by ferrocyanide of potassium, but it is open to so many objections that the process by weight is always preferred.

2. *Sugar.*—The variety of sugar found in urine is grape sugar $C_6H_{12}O_6$ frequently termed diabetic sugar. Brücke has demonstrated that sugar in small quantity may frequently be found in healthy urine ; but its constant presence in large amount in urine constitutes the disease known as diabetes (διὰ, through, and βαίνω, I pass). The urine in this disease is usually light coloured, froths readily on being poured from one vessel into another, and has a high specific gravity.

Preparation of diabetic sugar from urine.—Evaporate urine to consistence of syrup, and allow the sugar to crystallise out. It is still impure, being mixed with urea and extractive matters. Separate these by means of absolute alcohol, and then add to the residue spirits of wine which will dissolve the sugar. It is again allowed to separate out from this solution, and the crystal-

line masses purified from alcohol by repeated re-crystallisations from water. When thus obtained, it is white, and crystallises in little lumps. These are composed of crystals belonging to the rhombic system.

Tests for sugar in urine. (*a.*) *Moore's test with caustic potash.*—To the suspected urine add an equal bulk of solution of caustic potash, and boil. If sugar be present, a deep orange-brown (like dark sherry) will be obtained. If sugar be present in large quantity, the colour is dark purple, and frequently almost black. This colour is produced by the action of KHO on $C_6H_{12}O_6$ producing melassic and glucic acids. The caustic potash used should be freshly prepared, because if allowed to stand for a length of time in a glass bottle, it becomes contaminated with lead, which, acting on the sulphur of urine, produces black sulphide of lead, and gives rise to a deceptive colour.

(*b.*) *Trommer's test with sulphate of copper and caustic potash.* To the urine add a few drops of solution of sulphate of copper. To this add a little caustic potash. This throws down a greenish blue precipitate of hydrated cupric oxide ($CuOH_2O$), which is dissolved in excess of the caustic potash, forming a blue liquid. Heat this by applying the flame of the lamp to the upper stratum of the fluid, and if sugar be present, a yellow, or orange, or red precipitate of cuprous oxide (Cu_2O) will be formed, which will form a marked contrast to the blue liquid in the bottom of the test tube. This test depends on the fact that diabetic sugar has the property of reducing cupric oxide to cuprous oxide. It does not do so directly, but indirectly, by its decomposition by the action of caustic potash, into melassic acid, which has a strong tendency to unite with oxygen. Unfortunately, however, other substances, such as excess of urates, or the protein compounds occasionally present in urine, have the same property, especially with the assistance of prolonged boiling, and it is consequently often difficult to detect minute traces by means of this test. If the cupric oxide be reduced to cuprous oxide *in the cold*, we may be sure diabetic sugar is present.

(*c.*) *Fehling's test with potassio-cupric tartrate* ($K_2Cu2C_4H_4O_6$).—The composition and mode of preparing this solution will be subsequently described when treating of the volumetrical estimation of sugar (p. 482). A few drops of it are added to the urine, and the upper stratum boiled. If sugar be present, it will reduce the cupric oxide in the alkaline tartrate to cuprous oxide, and

give the same reaction as in Trommer's test. If freshly prepared, Fehling's solution will often detect minute traces of sugar, but it is liable to decomposition if kept for even a week, and occasionally it gives uncertain results, even when the presence of sugar has been ascertained by the other tests.

(*d.*) *Böttcher's test with nitrate of bismuth.*—Add to the urine an equal volume of a solution of carbonate of soda (3 parts of water to 1 part of crystallised Na_2CO_3), and afterwards a little trisnitrate of bismuth, and boil. If the white powder become dark, sugar is present, owing to the fact that sugar has the power of reducing the oxide of bismuth. If albumin be present in the urine, it must be first got rid of by boiling and filtration, because the sulphur of the albumin may readily form with the bismuth black sulphide of bismuth.

(*e.*) *Dichloride of tin test.*—Moisten a few strips of merino in a solution of stannous-chloride, and dry in a water bath. On moistening one of these strips with diabetic urine, and holding it near the fire, a brownish-black colour will make its appearance.

(*f.*) *Fermentation test.*—Ordinary yeast is mixed with water, and a long test-tube filled with the suspected urine, to which some of the yeast has been added. The tube is then inverted over a saucer containing the urine under examination, so that no air may enter, and the whole is set aside in a warm place. If sugar be present, it will be decomposed under the action of the yeast into carbonic acid and alcohol, and the gas will speedily collect in the upper part of the tube. Another mode of demonstrating the change is to conduct off the carbonic acid by a fine tube into lime-water, which of course at once becomes turbid from the formation of insoluble carbonate of lime.

Estimation of the amount of sugar.—This may be done in two ways: (1.) by a volumetrical process; and (2.) by means of an instrument termed a saccharimeter.

1. *Volumetric process for Diabetic Sugar.*

This process is founded on the property already mentioned which diabetic sugar possesses of reducing cupric oxide to cuprous oxide. If we use, therefore, a solution of potassio-cupric tartrate which contains, in a given volume, a quantity of cupric oxide that is reduced by a certain quantity of sugar, we can estimate the amount of sugar in solutions of unknown

strength by finding the volume required for the decomposition of a fixed quantity of copper solution.

Preparation of the copper solution (*Fehling's solution*).—34·65 grammes of pure crystallised sulphate of copper are dissolved in about 160 grammes of water; and a solution of 173 grammes of pure crystallised double tartrate of potash and soda is treated with from 600 to 700 grammes of caustic potash of 1·12 sp. gr. Into the latter solution the sulphate of copper solution is gradually poured. The clear mixture is then diluted up to a litre. It has been found that 10 C. C. of this copper solution are reduced by exactly 0·05 gramme of diabetic sugar. In order to preserve the copper solution for a length of time, it is necessary to keep it in a dark place, in small stoppered glass bottles (containing 1 or 2 ounces).

Process.—(1.) Filter the urine.

(2.) Dilute the urine with 20 times its bulk of distilled water, and place it in a burette.

(3.) Dilute 10 C. C. of standard solution (=·05 grammes of sugar) with 20 to 30 parts of distilled water, and place it in a porcelain capsule, under the burette.

(4.) Boil, gradually adding the diluted urine from the burette, until the cuprous oxide has been precipitated as a reddish powder, and the supernatant liquid has acquired a straw-yellow colour, not a trace of blue remaining.

(5.) Filter the *boiling* fluid, and divide into three portions :

a. 1st portion.—Add a few drops of hydrochloric acid, and afterwards a little of a solution of sulphuretted hydrogen. The absence of a black colour indicates that all the cupric oxide has been reduced.

b. 2nd portion.—Add a few drops of acetic acid, and then test with ferrocyanide of potassium. The absence of a reddish brown colour or precipitate indicates that all the cupric oxide has been reduced.

c. 3rd portion.—To guard against the error of adding too much urine, add to this portion a few drops of the copper solution, and boil. If a trace of sugar be present, a reddish colour will appear in a short time.

Example—Patient passes 15,000 C. C. of urine, 36 C. C. of dilute urine were required to reduce all the cupric into cuprous oxide in 10 C. C. of standard solution. How much sugar was passed? 10 C. C. of S. S. require exactly ·05 grammes of sugar

to effect the reduction. 36 C. C. of dilute urine=1·8 C. C. of real urine (20 : 1 : : 36 : 1·8) .·. 1·8 C. C. of real urine contain ·05 gramme of sugar. Then 1·8 : ·05 : : 15,000 : 416·6̇ grammes of sugar in 15,000 C. C. of urine.

2. *Estimation of Sugar by the Saccharimeter.*

It is well known that diabetic sugar, in common with cane sugar, milk sugar, camphor, &c., have the property of rotating the plane of the vibrations of a ray of polarised light to the right (p. 142). It has been found also that the angle of deviation is in proportion to the length of the column through which the ray passes, or to the quantity of the substances contained in a column of given length. The saccharimeter of Soleil consists essentially of four parts : 1. A glass tube, for containing the fluid to be examined, fitted into a brass case, and closed at both ends with plate-glass discs ground to fit water-tight, and kept tightly in their place by means of screw-caps. This is placed on a support between a polarising and analysing apparatus. 2. A polarising apparatus consisting of an achromatic calc-spar prism, having a small screen behind it (that is nearer the tube containing the solution) which intercepts one of the images ; and a double plate of quartz, one half being dextro,—and the other lævro-rotatory. 3. An analysing apparatus, consisting of a quartz plate cut perpendicularly to its axis, and a doubly refracting prism, fitted into a small telescope ; and, 4. An apparatus, termed a *compensator*, which is placed between the quartz plate and doubly refracting prism just mentioned. This consists of two elongated quartz prisms cut perpendicular to the axis, each one being narrower at one end than at the other, and set on two racks moved horizontally by a toothed pinion, so as to vary the thickness which the modified light has to traverse. One rack carries a scale (tenths of a milimetre), the other a vernier (tenth of the tenth of a milimetre=1-100th of a milimetre) so as to measure the displacements of the prisms.

Mode of using the saccharimeter.—The instrument is placed before a bright light, the polarising apparatus being next the light. The tube is filled with distilled water, care being taken to exclude all bubbles of air, and is placed between the polarising and analysing apparatus. The eye is now directed to the telescope. If the instrument be correct, a disc of coloured light

is seen, divided into two hemispheres by a very faint dark line, when the zeros of the vernier and scale coincide. The tube is now emptied of distilled water, carefully dried, and filled with urine which has been prepared by adding to it a solution of acetate of lead, and filtering. When the polarised light is allowed to pass through this stratum of urine containing diabetic sugar, the two hemispheres will now be found to have different colours, say one red and the other blue, and the object is now to bring back the two hemispheres to the same tint by moving the compensating prisms, which is done by turning a screw attached to the pinion already mentioned. By thus moving the compensator, we produce an inversion of the rotation of the ray of polarised light opposite to that produced by the liquid, and the displacement of the vernier gives the angle of deviation—the thickness of quartz corresponding to one division of the scale being known. The number of degrees on the scale is now read off, and each degree corresponds to a certain amount of sugar in a known quantity of urine. The instrument in use in the physiological laboratory of Edinburgh University, is so adjusted that each degree of the scale corresponds to ·111 ounces of diabetic sugar in 50 ounces of urine. Thus, suppose a patient passes 200 ounces of urine in 24 hours, How much sugar is excreted? The urine is examined as above, and it is found that the zero of the vernier is opposite 28 of the scale when the tints of the hemispheres are exactly the same. Then $28 \times ·111 = 3·108$ oz. in 50 oz. $\therefore$ $3·108 \times 4 = 12·432$ oz. in 200 oz of sugar.*

3. *Bile.*—Urine containing bile has a peculiar greenish-black colour. The tests for bile acids and bile pigment have already been described at p. 458, while treating of bile.

4. *Fat.*—Occasionally fat is found in the urine in the form of oil globules, but it is usually associated with fatty casts, indicating an advanced condition of Bright's disease.

5. *Chylous urine.*—This urine is white, from the abundance of fatty molecules it contains. Sometimes albumin is present when it coagulates on cooling. It is probable in these cases there may be some abnormal communication between the lacteal system and the ureters or kidney.

6. *Kiestein.*—The urine of pregnant women often shews a fat-

* For a complete description and figure of the saccharimeter, see Watt's Dictionary of Chemistry—article, "Light." Vol. iii. p. 674. Also Desplats et Garièl's Nouveau Eléménts de Physique Médicale. Paris, 1870, p. 396.

like scum on the surface, which consists of crystals of triple phosphate, fat globules, and a granular matter of an albuminous nature called kiestein. When kept, it smells like old cheese.

7. *Lactic acid.*—This acid is rarely found in urine, and its presence cannot be determined by any special test, but by the following mode of procedure: Evaporate fresh urine nearly to dryness, and treat the residue with a solution of oxalic acid in alcohol. Oxalates are thrown down, while the lactic acid remains in solution. This fluid is then digested with litharge, evaporated to dryness, and an alcoholic solution of lactate of lead obtained. This, in turn, is decomposed by sulphuretted hydrogen, the sulphide of lead filtered off, and the fluid evaporated to a syrup. The syrup is now shaken up with ether, the etherial solution of lactic acid evaporated, and the lactic acid dissolved in water. The aqueous solution is now boiled with zinc oxide and the crystals of lactate of zinc are allowed to separate.

8. *Acetic and butyric acids.*—These are found only in decomposing urine, and it is not important to detect or isolate them.

9. *Sulphuretted hydrogen.*—This gas has rarely been found in urine. It may be readily detected by blackening a piece of paper dipped in a solution of acetate of lead and held over it.

10. *Allantoin* (Pl. I. fig. 8).—Schottin has found this substance in the urine of a man who had taken a large quantity of tannic acid. It has also been found in the urine of young children, but it is probable its presence is only temporary. Its detection is of no practical importance.

11. *Leucin* (Pl. I. fig. 10).—This product has been found in the urine of individuals suffering from hepatic disorders. There is no chemical test for its presence, and it can only be identified in deposits by microscopical examination. It usually is found in the form of roundish, yellowish coloured balls, which consist in reality of masses of small needle-like crystals.

12. *Tyrosin* (Pl. I. fig. 9).—It is formed under the same conditions as favour the production of leucin, and like it, can be identified only by means of the microscope. It consists of stellate groups of long silky needles, not in balls or coloured, as is the case with leucin.

Examination of the Sediments of Urine.

These may be conveniently divided into 1. those occurring

phates, or both, it may be distinguished from them by heating and adding acetic acid. The heating dissolves the urates, and the acid dissolves the phosphates, but neither have any effect on cystin.

2. *Deposits found occasionally in alkaline urine only.*

The formation of these has already been explained at p. 262 and p. 460. They are all dissolved on adding a few drops of nitric or hydrochloric acids. They are,

(1.) *Ammoniaco-magnesian, or triple phosphate* (Plate I. fig. 18).—This salt always exists in ammoniacal urine, and is easily recognised by its well-known crystalline forms. It is usually found in variously modified six-sided crystals, some elongated (A), others nearly square (see to right of *b*), some having sharp angles, while others have broad facets (*a*), and in very alkaline urine they appear as feathery crystals (*c*).

(2.) *Phosphate of lime.*— It is usually an amorphous white powder, but occasionally it appears aggregated into rosette-like crystals.

(3.) *Urate of ammonia* is always present in alkaline, and rarely in acid urine. It has been described above.

(4.) *Urate of lime* is also occasionally found in alkaline urine.

3. *Organised deposits.*

These are mucus, blood, pus, tube casts, spermatozoids, torulæ, sarcinæ, bacteria, vibriones, &c.

(1.) *Mucus.*—When urine is left at rest, cloudy transparent floculi are seen, which consist of mucus entangling various forms of epithelial cells, derived from the urinary passages. If the supernatant liquid be carefully poured off, and acetic acid added to the mucus, it coagulates, forming delicate molecular fibres (Plate IV. fig. 1).

(2.) *Blood.*—Urine containing blood has a peculiar smoky colour that the practised eye can readily detect, but the best test is to identify the blood corpuscles by means of the microscope. As a rule, the blood corpuscles are colourless and have lost their biconcave form, and are globular from the imbibition of water. Urine containing blood always contains a trace of albumin.

(3.) *Pus.*—If there be a thickish yellow deposit at the bottom of the vessel, which has a stringy consistence, it usually consists

of mucus containing pus. Pour off the supernatant fluid, and add to the deposit an equal bulk of caustic potash. It at once gelatinizes, becoming so thick and tough that it cannot be poured from the test tube. When pus is present in small quantity, by means of the microscope we can readily detect the pus corpuscles (Plate III. figs. 17 and 18).

(4.) *Tube casts.*—These bodies are detected by allowing any sediment to fall to the bottom of a conical glass, removing a small portion of it with a fine pipette, placing a drop on a slide, covering it with a thin glass, and examining it with a power of 250 diam. linear. Tube casts are of various kinds, but they may be conveniently classified under the following: *a.* Fibrinous casts, often containing blood discs; *b.* Desquamative casts, containing epithelial cells; *c.* Granular or fatty casts, containing numerous oil globules, free, or in the epithelial cells (Pl. XIII. figs. 12 and 13); *d.* Hyaline or waxy casts, solid and transparent, or containing epithelial cells, granules, and free nuclei.

(5.) *Spermatozoids, torulæ, sarcinæ, bacteria, vibriones, &c.*—These, occasionally found in urine, may all be readily detected by their characteristic microscopical appearance.

The gases of the urine are not of importance, and it is sufficient to state they are the same as those of the blood, and in variable proportion.

CLINICAL EXAMINATION OF THE URINE.

The examination of a specimen of urine is to be made in the following manner :—

1. *Colour*, whether pale from being dilute, dark from being concentrated, dark or greenish from presence of bile, smoky from blood.

2. *Smell.*—Fragrant from the existence of cystine, or sugar, &c., or fœtid from alkalinity.

3. *Measure quantity passed in* 24 *hours*, and observe whether there is excess or diminution.

4. *Specific gravity.*—Take the specific gravity, if possible, of the mixed urine. Normal sp. gr. 1020. If high, suspect sugar; if low, suspect albumin.

5. *Reaction.*—If acid, is it normally so or not? If excessively acid, examine for crystals of uric acid. If alkaline, ascertain whether the alkali is fixed or volatile.

6. *Heat.*—Heat a portion in a test-tube. If a precipitate appear, it may be albumin or phosphates. Add a drop or two of nitric or hydrochloric acids. If precipitate dissolve, *phosphates;* if not, *albumin.* If a deposit disappear on heating, we have *urates.* If it do not disappear,

add a drop of nitric acid. If now dissolved, we have *phosphates;* if not, *cystin.*

7. *Bile.*—Test for bile pigment and bile acids, if necessary (p. 458).

8. *Sugar.*—Test for sugar, if necessary (p. 481).

9. *Chlorides.*—Add a drop of nitric acid, and then nitrate of silver, *till a precipitate ceases to form.* Thus estimate the amount of *chlorides.*

10. *Microscope.*—Examine for blood, pus, cystin, oxalate of lime, leucin, tyrosin, tube casts, &c., by the microscope.

Analysis of the Fæces.

As may be expected, the constitution of this excretion varies considerably from time to time. There are always present fragments of the undigested remnants of food, fatty matter, fatty acids, bile pigment, and soluble salts, chiefly alkaline phosphates, the ammoniaco-magnesian or triple phosphate, with traces of sulphates and earthy phosphates. Undigested fragments are readily separated by suspending them in water; fatty matter may be taken up with ether and alcohol; and the ash obtained by incineration will yield the mineral ingredients. Dr Marcet states* that healthy human fæces contain an acid, *excretolic acid*, and a substance called *excretin*, both of which are soluble in ether. He obtains these substances by making an alcoholic extract of fæces. This deposits, after long standing, an "olive-coloured" acid, *excretolic acid.* The alcoholic solution is then treated with milk of lime which throws down *excretin*, with other substances. The excretin is now separated from these by ether. It is probable that these substances may not be fixed compounds. (See pp. 271–2.)

III. GENERAL QUALITATIVE EXAMINATION OF AN ANIMAL SOLID.

The analysis of tissues and organs is attended with even greater difficulty than in the case of animal fluids. It is important, if trustworthy results are desired, to operate upon at least 12 or 15 lbs. of the tissue. The following is the mode of procedure to be adopted :—†

1. Cut the tissue into small fragments, and allow it to macerate in cold water. Filter. Add to the filtrate a concentrated solution of barium hydrate to throw down *phosphates*, *sulphates*, *uric acid*, and *hypoxanthin.*

* Marcet, Phil. Trans. 1854, p. 265; and 1857, 403.

† Watt's Dictionary of Chemistry, vol. i. p. 252.

2. Filter again and evaporate to a syrup. During this operation, a film will collect on the surface, consisting probably of *barium carbonate* or *magnesium phosphate*, and possibly of *uric acid* and *hypoxanthin*.

3. Allow crystals to separate out of the syrup, and probably they will consist of *creatin*.

4. Extract the mother liquid with alcohol and ether, and thus obtain *lactates of potash and soda*, *inosite*, *creatinin*, and *leucin*.

5. Make a fresh extract by steeping a portion of the tissue in cold water. Boil, and *albuminous matters* will coagulate, and are to be separated by filtration.

6. Evaporate the filtrate to a syrup, and masses having a crystalline appearance may separate out. These consist of *leucin*. *Tyrosin* sometimes appears in the form of star-shaped groups of slender needles, which are insoluble in alcohol.

7. The mother liquids from the last-mentioned deposits contain *volatile acids*, *lactic acids*, &c.

8. Incinerate a known weight of the substance, weigh the ash, dissolve it in a little hydrochloric acid, and test the solution for *inorganic acids* and *bases*.

Thus we obtain a general knowledge of the chemical constituents of the tissue under examination. Special processes are requisite for special tissues.

IV. QUALITATIVE ANALYSIS OF SPECIAL ANIMAL SOLIDS.

Under this head we shall treat of the analysis of muscle, white fibrous tissue, yellow elastic tissue, tooth, cartilage, bone, the nervous system, and lastly, of liver.

Analysis of Muscle.

1. *Reaction.*—When quiescent muscle is tested with litmus paper, it is found to be neutral or slightly alkaline, but if the muscle be thrown for sometime into a state of tetanus by an interrupted current of electricity, it is found to become acid. This is generally supposed to be due to the formation of sarcolactic acid.

2. *Kühne's method of obtaining muscle-plasma.*—Kill two frogs, and inject into the blood vessels a weak solution of common

salt (1 per cent.), until all blood is removed. Then cut off all the muscle of the limbs, reduce it to fragments, and subject it to powerful pressure. A liquid is thus obtained, termed by Kühne, *muscle-plasma*, which soon coagulates, resolving itself into a clot, called muscle clot or *myosin*, and a fluid, *muscle-serum*.

3. *Examination of muscle-serum.*—If this fluid be obtained in sufficient quantity, it will be found to contain three modifications of albumin, each coagulating at a different temperature. When a portion of muscle-serum is heated to 30° C. a coagulation takes place ; increase the heat to 45° C. and there is a further coagulation ; continue heating until 75° C. and another large amount of albumin will fall down. Muscle-serum also contains many excrementitious substances, resulting from the retrograde change of the tissues (*Secondary Digestion*, p. 239), such as creatin, creatinin, leucin, tyrosin, urea, uric acid, &c. These are to be distinguished and separated by the special processes and tests already fully described.

4. *Examination of muscle-clot or myosin* (p. 10).—This will be found to be insoluble in water, ether, or alcohol, but it is very soluble in dilute acids or dilute alkalies, and especially so in a ten per cent. solution of common salt. If the common salt solution be added to distilled water, the myosin falls as a flaky precipitate.

5. *Syntonin* (p. 10).—Dissolve a portion of muscle-clot in a little weak hydrochloric acid. When this acid solution is added to water, a flaky precipitate is obtained, which is insoluble in a ten per cent. solution of common salt. This substance, which thus does not exhibit the characteristic reaction of myosin, has been termed by Kühne *Syntonin*. He holds that syntonin does not exist as such in muscle, but is an artificial product obtained by the action of dilute acid on myosin. Syntonin may be prepared from muscle in the following way :—Mince a piece of muscle and allow it to macerate in cold water until the water does not coagulate on boiling, shewing the absence of albumin. The macerated muscle is now treated with ten times its bulk of weak hydrochloric acid (·1 per cent.) and left to stand for 24 hours. Neutralise with carbonate of soda, and a white and gelatinous precipitate will fall, consisting of syntonin.

6. *Inosite.*—The muscular substance of the heart contains a peculiar saccharine substance, isomeric with glucose, $C_6H_{12}O_6$,

which may be separated as follows :—Macerate the heart in water, precipitate the phosphates with baryta-water, filter, evaporate the filtrate, and allow creatin to separate out. Treat the mother liquid with dilute sulphuric acid, which will precipitate the baryta. Filter so as to remove the sulphate of barium. Shake up the liquid with ether so long as anything is dissolved. Separate the ether by skimming, and mix with alcohol until a precipitate appears. This precipitate is sulphate of potash, which is now separated by carefully pouring off the supernatant fluid. Mix this latter with more alcohol, and soon small oblique or tabular prisms of inosite will separate (p. 27).

*Scherer's test for inosite.**—The following is a test for the presence of inosite. To an aqueous solution, evaporated nearly to dryness, add a drop or two of nitric acid, moisten the residue with a few drops of ammonia and calcium chloride, again evaporate, and a rose-coloured substance remains.

Incineration—Incinerate a given weight of muscle, and weigh the ash. Dissolve this in hydrochloric acid, and test for salts in the ordinary way.†

Analysis of White Fibrous Tissue.

1. *Basis of white fibrous tissue, gelatin.*—This tissue shrinks much on being dried. When allowed to macerate in water, or when boiled in water, a gelatinous mass is obtained, consisting of gelatin (p. 10). The jelly dissolves in hot water, and from the solution, alcohol precipitates a white clotted mass.

2. *Incineration.*—White fibrous tissue contains a very small amount of inorganic matter, which can be obtained by incineration.

Analysis of Yellow Elastic Tissue.

1. *Basis of yellow elastic tissue, elastin.*—Boil a piece of the *ligamentum nuchæ* of an ox with alcohol, then with water containing ten per cent. of strong hydrochloric acid ; allow it to cool, and a yellowish, fibrous, and brittle mass is obtained, termed elastin. This substance is insoluble in water, alcohol, ether, and acetic acid. It is dissolved by strong caustic potash.

2. *Incineration.*—There are more inorganic substances present

* Scherer, Ann. Ch. Pharm., lxiii. 322, lxxxi. 375.

† For details as to the chemical composition of "Flesh," or butcher meat, see Watt's Dictionary of Chemistry, vol. ii. p. 661.

in elastic tissue than in white fibrous tissue. The amount of these may be determined by incineration.

Analysis of Tooth.

1. *Separation of organic basis.*—By allowing fragments of teeth to macerate for three or four weeks in dilute hydrochloric acid (1 to 19 of water), a soft substance remains, which is probably gelatin. This constitutes the organic basis of teeth, and is present in larger quantity in dentine than in enamel.

2. *Incineration.*—By incinerating teeth, the organic matter is burnt off, and the ash will be found to consist chiefly of calcium phosphate, along with a much smaller quantity of calcium carbonate. Phosphate of magnesia, and a very minute trace of calcium fluoride, are also present. For examples of analyses of teeth, see p. 87.

Analysis of Cartilage and Bone.

1. *Water in cartilage.*—Weigh a piece of cartilage, allow it to dry in a hot-air chamber, and it will be found to have lost half its original weight. This is owing to the fact that cartilage consists largely of water.

2. *Preparation of chondrin* (p. 11).—Boil a few of the cartilages of the ribs or joints with water for 48 hours, evaporate to a jelly, and wash the jelly with ether to free it from fat. The jelly is chondrin. The various reactions of chondrin may now be demonstrated. It is soluble in boiling water, but insoluble in alcohol or ether. When any of the mineral acids are added to an aqueous solution, a precipitate is formed, which is redissolved in excess ; but the precipitate formed by carbonic, sulphurous, acetic, or tartaric acids, is not redissolved in excess. It should also be compared with gelatin, as follows :—

Reagent.	Chondrin.	Gelatin.
Alum.	Precipitate.	No precipitate.
Acetate of lead.	Precipitate.	No precipitate.
Sulphate of iron.	Precipitate.	No precipitate.
Mercuric chloride.	No precipitate.	Precipitate.

3. *Bone. Organic basis of bone, ossein.*—Allow fragments of bone to macerate for some time in dilute hydrochloric acid (1 to 19 of water). The calcium salts are dissolved, and a soft translucent mass remains, termed bone-cartilage or *ossein.* This substance resembles gelatin, but it differs from it in being

insoluble in boiling water. By prolonged boiling, however, ossein is converted into gelatin.

4. *Inorganic salts of bone.*—The acid solution of bone salts may now be evaporated, and the examination of these inorganic salts conducted in the usual method.

5. *Incineration.*—First reduce the bone to fine powder, wash with water to remove soluble salts, and with ether to remove fat. The powder is now incinerated (best of all in a muffle) till it becomes white. A few drops of solution of carbonate of ammonia are now added to it to make up for the loss of any carbonic acid driven off from the carbonate of lime present in bone, and it is again incinerated. The difference between the weights before and after ignition gives the amount of ossein. The ash is now to be analysed in the ordinary way.

The results of numerous analyses of cartilage and bone will be found at pp. 90, 92, 93.*

Analysis of the Nervous System.

The chemical composition of nervous tissue is still very imperfectly understood, and no definite mode of chemical analysis can be recommended.

1. *Water and fat.*—The amount of water may be ascertained by drying a certain definite weight, and will be represented by the loss in weight. The white matter of the nervous centres contains less water than the grey matter ; while the white, on the other hand, contains more fat than the grey matter. The amount of fat may be determined by acting upon nervous substances with ether.

Various physiological chemists have detected in nervous matter the following substances : leucin, uric acid, xanthin, inosite, creatin, creatinin, formic, and acetic acids.

2. *Cerebric acid.*—This is a fatty acid supposed to exist in the brain. Cut brain substance into thin slices, act upon it with boiling alcohol to remove water, press it, digest with cold, then with warm ether, distil off the ether, and digest with much more ether. We have now cerebrate of soda mixed with phosphate of lime, &c. Digest it in boiling absolute alcohol acidulated with sulphuric acid. We thus obtain an alcoholic solution of cerebric acid. When this is evaporated, the acid is deposited

* For analyses of bones of different animals, see article "Bone" in Watt's Dictionary of Chemistry, vol. i. page 619.

as a white crystalline substance. It is doubtful if this acid be a constant ingredient, and probably it is a substance produced artificially during the chemical process. Other substances have been found in brain, termed cerebrin, cerebrol, and cerebrote; but it is probable they are one and the same substance.

3. *Protagon.*—This is the name of a substance supposed to exist in brain. According to Liebreich, it is the principal constituent of nervous tissue. (*a.*) Reduce brain substance to a pulp, and act upon this with water and ether at 0° C. From the remaining substance extract the protagon by 85 per cent. alcohol at 45°. Cool the alcoholic solution to 0° C., and a precipitate is formed which, on being examined microscopically, is found to consist of bundles of crystals.

(*b.*) This substance may also be prepared in a somewhat different form from yoke of egg. Beat up very thoroughly the yoke of an egg in 2 ounces of absolute alcohol. Boil carefully and filter while hot. Allow the filtrate to drop upon a flat and cold porcelain plate, when a yellowish non-crystalline deposit will be found, consisting of protagon. The remarkable reactions of this substance are described at page 45.

4. *Oleo-phosphoric acid.*—This is a fatty acid found in the brain; it may be prepared as follows:—Beat up brain substance to a thin pulp with water, heat the mixture to the boiling point, and act on the coagulum formed with boiling alcohol. This extract is filtered while hot, and deposits cholestrin, cerebrin, and oleo-phosphoric acid, united with alkalies. Act upon this with cold ether, which takes up oleo-phosphate of soda. Evaporate the ethereal solution, decompose the oleo-phosphate by a few drops of dilute hydrochloric acid, dissolve the residue in boiling alcohol, and the oleo-phosphoric acid is deposited when it cools. It is a gummy or fatty yellowish substance, easily decomposed into phosphoric acid, and one of the higher fatty acids.

Analysis of the Liver.

It is well known that the liver contains a substance termed *glycogen*, isomeric with starch, $C_6H_{10}O_5$ (p. 25). It may be prepared from the liver, as follows: *Bernard's method.*—Cut a piece of liver into small portions, boil it for an hour in water, and allow the hot decoction to filter into glacial acetic acid. Nearly pure glycogen is thrown down, the albuminous substances remaining in solution. It is precipitated from its aqueous

solution by animal charcoal, and is usually quite insoluble in alcohol. It is important to observe that dilute mineral acids, diastase, and the peculiar nitrogenous ferments found in the blood, saliva, liver, and pancreas, readily convert glycogen into diabetic sugar, $C_6H_{12}O_6$. It is probable a change of this kind occurs with great rapidity on death, for we have found that a decoction of a liver removed from a rabbit or mouse just killed, always gives a characteristic reaction with any of the tests for sugar. If, however, a portion of the same liver be kept for several hours, and a decoction then made, a much more decided reaction will be obtained. (See p. 251.)

In conclusion, it may safely be asserted that chemical physiology is still in its infancy. It is a difficult field of labour, both from the complex constitution, as well as from the instability of many of the substances to be examined. Nor must we forget that the chemist can analyse only dead tissues and fluids, not living tissues, and many of the substances which are obtained by chemical processes in the laboratory do not exist as such in the living body.

PRACTICAL HISTOLOGICAL PHYSIOLOGY.

This subject can only be prosecuted with the aid of an achromatic microscope, the construction and mode of employment of which instrument must be first understood.

History of the Microscope.

A microscope (from *μικρός*, small, and *σκωπέω*, to see) may be defined, an instrument which is capable of making small objects appear larger than they do to the naked eye. In this sense, it applies to any instrument, of whatever contrivance, capable of fulfilling this condition ; and if we accept this definition, various reasons have been adduced to shew that the microscope was known to the ancients. Spectacles, it is said, were in use among the Greeks and Romans ; and as the glasses of these were made of different convexities, and, consequently, of different magnifying powers, it is natural to suppose that they must have been acquainted with the property possessed by the lens, of enlarging small objects. Various passages also occur in the works of

Jamblichus, Pliny, Plutarch, Seneca, and others, which lead to a similar conclusion. Thus, Seneca observes: "Letters, though minute and obscure, appear larger and clearer through a glass bubble filled with water." Now, this glass bubble filled with water is sold by pedlars and others to the vulgar at the present time, in order to magnify objects.

The compound microscope appears to have been constructed in the early part of the seventeenth century. Both Holland and Italy have claimed the honour of producing its inventor. William Borelli attributes its construction to one Zacharias Jansen of Middleburgh in the Low Countries, who, with his son John, according to this author, made his first compound microscope so early as 1590. It is stated that either he or his son presented one of his instruments to the Archduke Charles of Austria, who, in turn, gave it to Cornelius Drebbel, a Dutch alchemist, who subsequently became astronomer to James I. of England. He it was who first brought the instrument to London in 1619, where it was seen by William Borelli and other scientific individuals. It is well known that Drebbel made microscopes in London in 1621, and generally passed for their inventor.

On the other hand, Francis Fontana, a Neapolitan, states, that he invented the instrument in 1618, and gave a description of it in his "Novæ terrestrium et cælestium observationes." It would appear, however, that although Drebbel and Fontana disputed concerning the origin of this instrument, the honour of inventing it, so far as our present knowledge extends, belongs to Jansen.

The microscope brought by Drebbel to London is thus described by Adams, who observes: "It is possible that this instrument of Drebbel's was not strictly what is now meant by a microscope, but was rather a kind of microscopic telescope, something similar in principle to that lately described by Mr Æpinus in a letter to the Academy of Sciences at Petersburgh. It was formed of a copper tube, six feet long, and one inch in diameter, supported by three brass pillars in the shape of dolphins. These were fixed to a base of ebony, on which the objects to be viewed by the microsope were also placed."*

The improvement of the microscope made much less rapid progress than that of the telescope. The great utility of the

* Adams on the Microscope, p. 3.

latter, indeed, appears to have been early appreciated, while the microscope was for a long time only regarded as a means of satisfying curiosity. Thus it was merely looked upon as an expensive toy, and kept by the rich in their cabinets as a source of amusement. At a later period, however, it was found susceptible of adding much to our knowledge of the natural sciences ; and no sooner was this perceived, than the most celebrated artists, mechanics, geometricians, and natural philosophers paid great attention to its improvement. For a long time, however, they were baffled by the difficulties of the undertaking, and during this period naturalists, for the most part, employed the simple microscope. Thus, some of the most important discoveries in science have been made by means of a single biconvex lens, and the laborious and brilliant researches of Leuwenhoeck, Swammerdam, Lyonet, Ellis, and others were thus accomplished.

The inconveniences of the simple microscope, however, are considerable. Thus, when capable of magnifying largely, the field of vision is very limited, and there is great difficulty in adjusting the focus. Leuwenhoeck had a separate lens especially adapted to one or two objects, and always had several hundreds at his disposal.

The imperfections of the compound microscope, on the other hand, were at that time very great, and must have appeared insurmountable. Thus, from its peculiar construction, the rays of light were readily decomposed, and circles of different colours surrounded or tinged the object, constituting the aberration of refrangibility. The form of the object was also distorted on account of the aberration of sphericity. Opaque objects could not be seen from the absence of light, and very transparent ones could not be examined from its excess.

But gradually all these different obstacles were overcome by patience and labour. The details connected with these, however, we cannot enter into. Suffice it to say, that to Lieberkühn we are indebted for the means of examining opaque objects by means of a reflector ; to the diaphragm of Le Baillif, for a convenient mode of modifying an excess of light. Achromatic instruments were constructed principally through the ingenuity and labours of Euler, Dolland, Frauenhofer, Selligue, Amici, Tulley, and Vincent, and Charles Chevalier, and may be said to have been perfected only during the last thirty years.

The object of the optician at present engaged in manufacturing this instrument is to construct a microscope which will admit of an easy and universal application, and possess the power of magnifying largely, combined with clearness and distinctness of the image. The instruments now constructed by Ploesel in Vienna, Frauenhofer in Munich, Schiek of Berlin, Hartnach and Nachet in Paris, and Powell, Ross, and Smith in London, if they have not reached perfection, certainly approach very near it, and permit the most minute details of structure to be examined with ease, even when magnified largely.*

Optical Principles on which the Microscope is constructed.

The optical principle on which every microscope is constructed is, that rays of light passing through a lens are more or less refracted, that is, are bent out of the straight line. This has already been fully explained at page 135.

Theory of enlargement.—The theory of a simple bi-convex lens will be understood by referring to Plate XXI. fig. 3. Here we have a convex lens interposed between the eye and a small object $a\,b$. If $a\,b$ be very close to the eye, the rays passing from it would diverge so far that the optical arrangements of the eye itself would fail to bring them to a focus on the retina, because the eye is adapted to receive and bring to a focus rays which are parallel or but slightly divergent. But when the lens $x\,y$ is placed between the eye and the object, the rays $a\,x$, $b\,y$, are so refracted by the lens as to come to a focus on the retina. Thus a well-defined picture or image is formed. But the rays now enter the eye at a greatly increased angle, and, consequently, the small object $a\,b$ appears increased in size to $a'\,b'$. It will be evident also, that if the lens $x\,y$ were more convex the effect would be further increased, as the refraction would be greater, and the object $a\,b$ would be seen still larger than $a'\,b'$.

* To no one is science more deeply indebted than to the late Mr Oberhæuser of Paris. He it was who first made good microscopes *cheap*, and brought them within the reach of the poorest scientific cultivator. Thousands of his instruments have been scattered over the world, and by their aid most of the facts on which the science of histology is founded were discovered. His nephew, M. Hartnach, continues his system with the like success.

A simple lens is termed a *simple microscope.* The same theory applies to increased convexity of the lenses in the eye-piece, only it is the image which is transmitted by the objective (p. 507) that is then magnified. It follows that any imperfection it possesses will be magnified also, so that the excellence of the objective is always the chief consideration in obtaining magnifying power. A third method of obtaining enlargement is by elongation of the tube, which, by causing greater divergence of the rays, also increases the apparent size of the object.

Faults of simple lenses and their corrections.—Every simple lens has two faults or optical imperfections—1st, that of spherical aberration, and 2nd, that of chromatic aberration. The modes of remedying these faults cannot be understood without an acquaintance with their causes.

Spherical aberration. (See p. 137.)—By referring to Plate XXI. fig. 4, it will be seen that all the rays of light passing through a convex lens do not come to the same focus in consequence of the refraction being necessarily greater at the circumference than towards the centre. Thus the rays *a* and *c*, as they impinge upon the glass at a greater angle, come to a focus at A, while the rays *b*, which are nearer the centre, come to a focus at B. This is owing to the unequal refraction of the rays, the rays *a* and *b*, passing through the margin of the lens being more refracted than those of *b* passing through its centre. Consequently, an image formed on the retina either at A or B would not only be indistinct and imperfect, but curved according to the convexity of the lens. There are various methods adopted for correcting spherical aberration.

1. By using a double convex lens the radii of which are as 1 to 6, with its most convex face turned towards the object.

2. By using a stop in the eye-piece, which is a plate with a round aperture interposed between the lens and the eye (Fig. 1, *c*), so as to cut off the rays *a c* (Fig. 4), and receive only those coming from *b*. Such an arrangement is used in all compound microscopes.

3. By using combinations of lenses, so disposed that the aberration of the one will correct the aberration of the other. Thus the aberration of one plano-concave lens may be made to correct that of another, so that all the rays will be brought to one focus, as in the eye-piece of Huyghens. This arrangement

will be described after we have considered the other imperfection of simple lenses, namely :

Chromatic aberration.—When an object is examined by a simple lens it will be found surrounded by rings of colour—red, orange, yellow, and so on. This appearance arises from the fact that the lens acts as a prism (see p. 140), and decomposes or disperses the ray of white light into its constituent coloured rays—red, orange, yellow, green, blue, purple, and violet. The most refrangible of these rays are the violet, the least the red, while those between these two colours possess different degrees of refrangibility. On referring to Plate XXI. fig 2, it will be seen that the rays of white light *a x* and *c y* are decomposed into the violet coloured rays *x* A and *y* A, and into the red rays *x* T and *y* T, the intermediate coloured rays, purple, blue, green, yellow, and orange not being represented in the diagram. In consequence of the great refrangibility of the violet rays, they are brought to a focus at A, while the least refrangible rays, the red, meet at T, a point farther from the lens than A. If the retina were situated at A, a coloured image would be seen, the centre violet, then purple, blue, green, yellow, and orange, while the margin would be red. On the other hand, if the retina were at T, a coloured image having the centre red and the margin violet would be the result. These fringes of colour seriously interfere with a correct interpretation of microscopic appearances, and must be got rid of. This is effected

1. *By the compound acromatic lens.*—This consists essentially of a bi-convex lens of crown glass and a plano-concave lens of flint glass carefully adjusted and cemented together, as seen in Pl. XXI. fig. 1, *e*, *f*, *g*, and figs. 5, *a*, and 6. The principle is, that as the dispersive power of the flint glass is so much greater than that of the crown glass, the one exactly corrects the other, and we thus have dispersion destroyed without destroying the refraction. When this is accomplished, we have *achromatism*, or the refraction of light without decomposition. At the same time, the refractive power of the two kinds of glass being also so different as exactly to neutralise each other's defects, we remove spherical aberration by this arrangement of lenses, and we thus obtain a distinct image on the retina by all the rays being brought to a focus without dispersion. It has been found best to use a combination of three such double lenses, Fig. 1, *e*, *f*, *g*, and Fig. 5, *a*. In Hartnach's and Nachet's microscopes, each pair of these are

fitted into small rings of brass which are screwed the one before the other. In the objectives of the London makers, a section of one of which is shewn (Plate XXI. fig. 6), the front pair can be approximated to the other two pairs, or the distance between increased so as to adjust the lenses for examining objects with or without a covering glass.

Object glasses having this adjustment, are constructed as follows (Plate XXI. fig. 6). The two higher achromatic lenses are fixed in the end of the tube B ; upon this slides a cylinder AA, carrying at the lower end a third lens, which, by turning the screwed ring CC, may be approximated to, or separated from, the other two lenses. These lenses can thus be so adjusted that the positive aberration of the anterior lens corrects the negative aberration of the two posterior, and also the aberration produced by even a thin covering glass when one is used. This improvement we owe to Mr Ross, the eminent optician of London.

2. *The eye-piece of Huyghens.*—It consists (Pl. XXI. fig. 1, *c c*, *b d*) of two plano-convex lenses, *b d*, with their plane sides towards the eye. These are placed, with regard to each other, at a distance equal to half the sum of their focal lengths. The upper one, *b*, is termed the *eye-glass*, the lower, *d*, the *field-glass*. A stop or diaphragm is placed at *c c* in the visual focus of the eye-glass, which is the same position as that where the image produced by the field-glass *d* is formed. Huyghens made this arrangement of lenses to correct spherical aberration merely ; but Boscovitch shewed that it also corrected chromatic aberration. This correction is now completely attained in all good Huyghenian eye-pieces. The rays of light passing into the eye-piece by the margin of the convex surface of the field-glass *d*, are decomposed so as to form two coloured images near the position of the eye-glass, the upper one blue and the lower red. The eye-glass, in its turn, would then magnify these so as to produce two secondary-coloured images near *y x*. These coloured images are combined so as to form a colourless image *y x :* 1st, by using a stop *c c*, which intercepts the rays passing through the margin of the lens ; and 2d, by having the eye-glass slightly over corrected for chromatic aberration, so that its focus would be shorter for blue rays than for red rays by just the difference in the place of the images formed at *y x*. Thus the rays enter the eye through the eye-glass in a parallel direction, and produce a picture free from colour.

Arrangement of lenses.—A section of a compound microscope is seen in Plate XXI. fig. 1. At the lower end of the tube there are one or more combinations of achromatic lenses, termed the *objective*, *e*, *f*, *g*, and at the other the eye-piece, *c c*, *b d*. An inverted image of *x y*, a small object placed under the objective, is made and inverted in the tube of the microscope in front of *d*, the field-glass; this image is magnified and again inverted by *d*, so as to form an image beneath *b*, the eye-glass, which last image is a third time inverted by the eye-glass *b*, which also directs the rays of light into the eye *a*, so as to form a distinct image on the retina. Thus the image on the retina is reversed as regards the object, a fact to be remembered in making microscopical observations.

Modes of increasing magnifying power.—This may be done in one or more of three ways: 1st, by increasing the length of the tube; 2d, by increasing the power of the eye-piece; and 3d, by using a higher objective. The objection common to all of these arrangements is, that while we increase the power we lose light. In the first instance we lose light by distributing it over a greater length of tube; in the second, we find that while we gain in power we lose brightness and definition, because any faults of the lens are of course intensified by the eye-piece. By using higher objectives, if they are good we obtain clearness of definition, with a sacrifice of light by dispersion of the rays. This, however, can be diminished by illumination, so constructing the lenses as to permit the passage of a large amount of light. Such a lens is said to have a large angle of aperture. (See Fig. 5^a, where *a b c* is the angle of aperture.)

All three modes of enlargement, viz., by the objective, by the eye-piece, and by elongation of the tube, are taken advantage of in the best instruments, and are useful within certain limits, as in the model I recommend, now manufactured by Hartnach.

Construction of the Microscope.

A microscope may be divided into mechanical and optical parts.

Mechanical parts.—These determine its general form and appearance. Of the numerous models which have been invented, the one figured (Pl. XXI. fig. 8), one-eighth its real size, appears to me the most useful for all the purposes of the physiologist and medical practitioner. It was suggested by me to the late Mr

Oberhæuser, and manufactured by him with his accustomed ingenuity. (*b.*) The body consists of a telescope tube, eight inches in length, held by a split tube (*c*), three inches long. It may be elevated and depressed with great readiness with a cork-screw movement, communicated to it by the hand, and this constitutes the coarse adjustment. It is attached to a cross bar and pillar, at the lower portion of which last, very conveniently placed for the hand of the observer, is the fine adjustment (*f*). The stage (*d*) is three inches broad, and two and a half inches deep, strong and solid, with a circular diaphragm below it. The height enables the observer to rest his two hands edge-ways on each side, and to manipulate objects on its surface with the thumbs and fore fingers. The base of the instrument is heavily loaded with lead to give it the necessary steadiness.

This form of microscope possesses all the mechanical qualities required in such an instrument. These are—1st, steadiness ; 2d, power of easy adjustment ; 3d, facility for observation and demonstration ; and 4th, portability.

1. *Steadiness.*—It must be evident that if the stage of the microscope possesses any sensible vibration, minute objects, when magnified highly, so far from being stationary, may be thrown altogether out of the field of view. Nothing contributes more to the comfort of an observer than this quality of a microscope, and great pains have been taken to produce it. In the large London instruments this end has been admirably attained (Plate XXI. fig. 7), but at so much cost and increase of bulk as to render it almost useless. In the small model I have recommended, all the steadiness required is present in the most convenient form.

2. *Power of easy adjustment.*—It is a matter of great importance to those who use the instrument much, and work with it for hours together, that the adjustments should work easily and rapidly, and be placed in convenient situations. Nothing can be more commodious than the manner in which these ends are arrived at in the model figured. By insertion of the body of the instrument (*b*) within a split tube (*c*), you may, by a spiral movement, elevate and depress it with the greatest rapidity, and even remove it altogether if necessary. The necessity of continually turning the large screws affixed to most microscopes (Fig. 7, D), becomes fatiguing in the extreme. Then the fine adjustment (*f*) placed conveniently behind the microscope,

near the hand which rests on the table, is in the very best position ; whereas, in some London instruments, it is placed on the top of the pillar, so that you must raise your hand and arm every time it is touched (Fig 7, F). In other London instruments, it is placed in front of the body, so that you must stretch out the arm and twist the wrist to get at it. No one could work long with so inconvenient a contrivance.

3. *Facility for observation and demonstration.*—For facility of observation and demonstration, it is necessary that the instrument should be of a convenient height, and that the stage on which the objects are placed should be easily accessible. Here, again, nothing can be more commodious than the microscope I have recommended, for, when it is placed on the table, its height is almost on a level with the eye, and we can look through it for hours without the slightest fatigue. On the other hand, the stage (*d*) is elevated just so much as enables the two hands, resting on their external edges, to manipulate with facility all kinds of objects placed upon it. The large London instruments are so high as to render it necessary to stand up to see through them. To obviate this disadvantage, a movement is given to the body, by which it can be depressed to any angle (Plate XXI. fig. 7). But this movement renders the stage oblique, and removes it to a distance, where it becomes very inconvenient to manipulate on its surface. To obviate this difficulty, the stage itself has been rendered moveable in various ways by different screws (Fig. 7), so that in this way complexity has been added to complexity, until a mass of brass work and screws is accumulated, to the advantage of the optician, but to the perplexity and fatigue of the observer. But by no contrivance is it possible to avoid the aching arms which such a position of the stage invariably produces in those who work with such a cumbrous machine for any length of time. Hartnach has recently placed a joint on his small microscopes, which, when they are bent, brings the eye-piece opposite the observer's stomach. Except for the purpose of drawing with a camera it is utterly useless.

4. *Portability.*—This is a property which should by no means be overlooked in instruments that are intended more for utility than ornament. A medical man is often called upon to verify facts in various places ; at his own house, at an hospital, at the bed-side of his patient, or at a private post-mortem examination. It is under such circumstances that the value of portability is

recognised. The large London instruments require an equipage or a porter to transport them from place to place ; even the putting them in and out of the large boxes or cabinets that are built around them, is a matter of labour. In short, notwithstanding the splendour of the screws, the glittering of the brass, and the fine workmanship (Plate XXI. fig. 7), there can be little doubt that, on the whole, they are very clumsy affairs.

There are many occasions on which a medical man may find it useful to carry a microscope with him, especially in the case of post-mortem examinations. Many attempts have been made to construct a pocket microscope ; and for the purposes above alluded to, I myself caused one to be constructed some years ago which, with its case, resembled a small pocket telescope. Dr Gruby of Paris, however, has planned the most ingenious instrument of this kind, which possesses most of the properties we have enumerated, and will be found very useful for those accustomed to microscopic manipulation. It is contained in a case the size of an ordinary snuff-box, and possesses all the conveniences of the larger instruments, with various lenses, a micrometer, slips of glass, needle, knife, and forceps in that small compass.* It is deficient in steadiness, however, a fault which has been removed by a similar instrument made by Nachet of Paris, the box of which is made of brass (Plate XXI. figs. 9 and 10), and which I can strongly recommend for its usefulness and excellence.

There is a general feeling among the public that the larger a microscope is, the more it must magnify ; but this is an error. A very imposing mass of brass work and mechanical complexity is no guarantee that you will see objects better, or what is of more consequence, become good observers. On the contrary, the more unwieldy the instrument, the less disposed will you be to use it. Besides, the habitual employment of artificial methods of moving about the object, as by the screws of a moveable stage, will prevent your acquiring that dexterous use of your fingers and accuracy of manipulation which are at all times so useful. Nothing, indeed, can be more amusing than to see a man twisting his screws, pushing his heavy awkward stage about, and laboriously wasting time to find a minute object which another can do in a moment, and without fatigue, by the

* For a representation of this instrument, see my Clinical Medicine, fifth edition, pp. 97 and 80.

simple use of his fingers. But perhaps you will consider the weightiest objection to the large instruments is the expense they necessitate,—the cost being necessarily in proportion to the amount of brass and mechanical labour employed upon them. If, then, you have to choose between a complex model and a simple one, I strongly advise you, as a matter of real economy, to choose the latter.

We have next to speak of the optical parts of microscopes, which are certainly much more important than the mechanical ones—for everything depends upon obtaining a clear and distinct image of the object examined. Under this head we may describe the objective, the eye-piece, and methods of illumination.

1. *The Objective, or series of Achromatic Lenses*, is that part of the optical portion of a microscope which is placed at the bottom of the tube or body, and is near the object to be examined. This may be considered the most important part of the instrument, and the greatest pains have been taken by all opticians in the manufacture of good lenses. It is here, I consider, that the London opticians are pre-eminent, for I am not aware that in any part of the world more perfect objectives have been manufactured than the eighth of an inch by Smith, the twelfth of an inch by Ross, and the sixteenth of an inch by Powell. The latter has also manufactured the twenty-fifth of an inch, which I have used with advantage. And Dr Beale tells us he has made for him a lense of one-fiftieth of an inch focus. But when we come down to one-fourth of an inch, which is by far the most useful objective for histological and medical purposes, the superiority of the London opticians is very slight, if any. At this magnifying power the compound lenses of Hartnach and Nachet of Paris; Schiek and Pistor of Berlin; Frauenhofer of Munich, and Ploesel of Vienna, may be employed with the greatest confidence, and it may be said that by far the largest number of important discoveries in science have been made through their employment. The Parisian lenses in addition, have one great advantage, namely, their cheapness.

The London opticians have succeeded in combining the lenses of their objectives so as to obtain a large field of vision, with as little loss of light as possible (Plate XXI. figs. 5, *a*, and 6). These qualities are valuable in the lower magnifying lenses during the examination of opaque objects, and in the higher

ones when observing transparent objects by transmitted light. But in the lenses of medium power, such as the one-fourth of an inch, those of Hartnach and of other continental makers are equally good.

In recent times so-called immersion lenses have been employed with advantage, that is, lenses so made that they may be depressed in a drop of water placed upon the covering glass. The object is thereby more highly illuminated and the focal distance increased. The highest powers can in this way be obtained at a much more reasonable price, from Hartnach, than from the London makers, and they are excellent.

For the above reasons, as well as from considerable experience in the use of many kinds of microscopes by different manufacturers, I am satisfied that the best lens you can employ for ordinary purposes is Hartnach's No. 7, which corresponds to what is called in England the quarter of an inch. For low powers you may have Hartnach's No. 3, or the one-inch lens of the London opticians. For all the wants of the medical man these will be sufficent. Occasionally the higher lenses may be required by the physiologist, as during the examination of the ultimate fibrillæ of muscle. These, by whoever made, may be attached to the model we have recommended by means of a brass screw made on purpose.

2. *The Eye-piece.*—This is that portion of the optical apparatus which is placed at the upper end of the tube or body, and is near the eye of the observer (Fig. 8, *a*). While the objective magnifies the object itself, the eye-piece only magnifies the image transmitted from below. Hence, as a source of magnifying power, it is inferior to the lens; and when this possesses any defects, these are enlarged by the eye-piece. Two eye-pieces are all that is necessary with the model I have recommended, and those of Oberhæuser, called Nos. 3 and 4, are the most useful for the medical man.

3. *Methods of illumination.*—There are few things of more importance to the practical histologist than the mode of illumination. This is accomplished—1st, by transmitted light; 2d, by direct light; and 3d, by achromatic light.

Transmitted light is obtained by means of a mirror placed below the object, which, to be seen, must therefore be transparent (Plate XXI. fig. 8, *e*). In large microscopes the mirrors are provided with universal joints, so that they may easily be

turned in any direction (Fig. 7, M). Below the stage every microscope should possess a diaphragm pierced with variously sized holes, whereby the amount of light furnished by the mirror may be moderated. In Oberhæuser's and Nachet's instruments the smallest aperture should be employed for the higher objective. It is also useful in the examination of many objects that the light should be directed upon them obliquely; this may be done by the diaphragm, or by the mirror, and in the small model (Fig. 8) is admirably attained by simply turning the whole microscope. The best light for microscopic purposes is that obtained by catching the rays which are reflected from a white cloud. The conjoined use of the mirror and diaphragm can only be learned from actual experience.

Direct light is employed in the examination of opaque objects, and the lenses of low power, manufactured by the principal London opticians, enable us to do so without assistance. Occasionally, however, the light of the sun is useful; and when this cannot be obtained, the rays of a lamp or gas light, concentrated by a bull's-eye lens, may be employed (Fig. 8, *g*). Hence every microscope should be possessed of such a lens, and it is most convenient to have it attached to the body of the instrument by a moveable ring, and stem with one or two joints, as in the model figured (Plate XXI. fig. 8).

Achromatic light is only serviceable in the examination of very delicate objects, with high powers. The apparatus necessary for obtaining it is occasionally useful in ascertaining the ultimate structure of muscle, or the nature of the markings on minute scales or fossils. The most elaborate instrument for obtaining condensed and achromatic light is Gillett's condenser (Fig. 7, *g*). It can only be adapted to the large instruments, and is seldom used. In the same way I know of little benefit to be obtained by a polarising apparatus.

In addition to the mechanical and optical parts constituting the microscope itself, the box which contains it should possess a convenient place for holding a few slips of glass, a pair of small forceps, a knife, and two needles firmly set in handles. A micrometer to measure objects with is also essential to those who are making observations with a view to their exact description. No other accessories are necessary.

An excellent microscope of the model figured (Fig. 8), by Hartnach, with two objectives (Nos. 3 and 7), two eye-pieces

(Nos. 3 and 4), a neat box with all the accessories necessary (with the exception of a micrometer, which had better be English) may be obtained in Paris for the sum of 140 francs (£5 12s.), and ought not to cost in Edinburgh, after payment of carriage, more than seven pounds. Nachet's instruments are somewhat cheaper. Either of them, for all the purposes of the student, is amply sufficient.

Test-objects.—The defining power of a microscope is generally tested by examining with it a transparent object, having certain fine markings, which can only be rendered clearly visible when the glasses are good. In all such cases, it is of course necessary to be familiar with the structure of the test-object in the first instance. If you are not confident on this point, it is better to trust to the judgment of a friend whose knowledge of histology is ascertained, or place your dependence entirely on a respectable optician. One of the best test-objects for a quarter of an inch lens is a drop of saliva from the mouth. For, if a microscope shews with clearness the epithelial scales, the structure of the salivary globules, their nuclei, and contained molcules, you may be satisfied that the instrument will exhibit all the facts with which, as medical men, you have to do. (See Plate IX. fig. 3.)

Mensuration and Demonstration.

Having obtained a good instrument, and tested its qualities in the manner described, the student should next determine the number of diameters linear the various combinations of glasses magnify. This he may do for himself with the aid of a micrometer, a pair of compasses, and a measure.

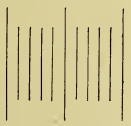
A micrometer is a piece of glass on which lines are ruled at the distance of $\frac{1}{100}$th or $\frac{1}{1000}$th of an inch. This must be placed under the instrument, when the lines and the distances between them will of course be magnified by the combination of glasses employed, like any other object. Taking a pair of compasses in one hand, we separate the points, and place them on the stage (always on a level with the micrometer magnified). Now, looking through the instrument with one eye, we regard the points of the compass with the other, and mark off by the naked sight, say the $\frac{1}{10}$th of an inch, as magnified by the instrument. Though difficult at first, a little practice enables us to do this with the greatest accuracy. The result is, that if the distance

magnified and so marked off ($\frac{1}{100}$th of an inch) is equal to three inches, the instrument magnifies 300 times linear; if two inches, 200 times; and so on.

To measure the size of objects, they may be placed directly on the micrometer; but as this is at all times inconvenient, whilst the object and micrometer, from their not being in the same plane, cannot, under high powers, both be brought into focus at once, it is better to use an eye-micrometer. Many ingenious inventions of this kind are to be procured. The most simple is a ruled micrometer placed in the focus of the upper glass of the eye-piece. With this we observe how many divisions of the eye-micrometer correspond with one of those magnified by the microscope, always making our observation in the centre of the field, where the aberration of sphericity is least. On the latter being removed and replaced by an object, it becomes a matter of mere calculation to determine its size. Thus, supposing each of the spaces in the upper to represent the $\frac{1}{1000}$ th of an inch magnified 250 diameters linear, and five of the lower spaces, as seen in an eye-micrometer, to correspond with one of these—it follows that each of these latter must measure $\frac{1}{5000}$ th of an inch.

Spaces equal to 1-1000th of an inch magnified 250 diameters linear.

If it be not in your power to estimate the magnifying power for yourself, the optician will construct a table, setting forth the various degrees of enlargement possessed by the lenses, and different eye-pieces with the tube up or down. This table should always be referred to during the description of objects, and the amount of magnifying power invariably stated.



Five ruled spaces in an eye-micrometer, corresponding to one of those above, and consequently equal to the 1-5000th of an inch.

The art of demonstrating under the microscope is only to be acquired by long practice, and like everything requiring practical skill, cannot be learnt from books or systematic lectures. I can only here give very general directions on this head.

All that is necessary in examining fluid substances, is to place a drop in the centre of a slip of glass, and letting a smaller and thinner piece of glass fall gently upon it, so as to exclude air

bubbles, place it upon the stage under the objective. In this way the fluid substance will be diffused equally over a flat surface, and evaporation prevented, which would dim the objective. The illumination must now be carefully arranged, and the focus obtained, first by means of the coarse, and then by means of the fine, adjustment. It will save much time, in examining structures, to employ always, at one sitting, the same slips of glass, as it is easier to clean these with a towel, after dipping them in water, than to be perpetually shifting the coarse adjustment.

The action of water, acetic acid, and of other re-agents, on the particles contained in a fluid, may be observed by mixing with it a drop of the re-agent before covering with the upper glass ; or if this be already done, the drop of re-agent may be placed at the edge of the upper glass, when it will be diffused through the fluid under examination by imbibition.

The mode of demonstrating solid substances will vary according as they are soft or hard, cellular or fibrous. The structure of a soft tissue, such as the kidneys, skin, cartilage, &c., is determined by making very minute, thin, and transparent slices of it in various directions, by means of a sharp knife or razor. These sections should be laid upon a slip of glass, then covered over, and slightly pressed flat, by means of an upper one. The addition of a drop of water renders the parts more clear, and facilitates the examination, although it should never be forgotten that most cell-structures are thereby enlarged or altered in shape from endosmosis. Acid and other re-agents may be applied in like manner. The double-bladed knife of Valentin will enable us to obtain large, thin, and equable sections of such tissues, and permit us to see the manner in which the various elements they contain are arranged with regard to each other. Harder tissues, such as wood, horn, indurated cuticle, &c., may also be examined after making thin sections of them. Very dense tissues, such as bone, teeth, shell, &c., require to be cut into thin sections, and afterwards ground down to the necessary thinness. Preparations of this kind are now manufactured on a large scale, and may be obtained at a trifling cost. A cellular parenchymatous structure, such as the liver, may be examined by crushing a minute portion between two glasses. If it be membranous, as the cuticle of plants, epithelial layers, &c., the membrane should be carefully laid flat upon the lower

glass, and covered with an upper one. The fibrous and tubular structures such as the areolar, elastic, muscular, and nervous tissues, must be separated by means of needles, and then spread out into a thin layer before examination, with or without water, &c.

The commencing observer should not be discouraged by the difficulties he will have to encounter in dissecting and displaying many tissues. He must remember that the figures he sees published in books are generally either fortunate or very carefully prepared specimens. Practice will soon enable him to obtain the necessary dexterity, and to convince himself of the importance of this mode of inquiry. He should early learn to draw the various objects he sees, before and after the action of re-agents, not only because such copies constitute the best notes he can keep, but because drawing necessitates a more careful and accurate examination of the objects themselves. A note-book and pencil for the purpose should be the invariable accompaniments of every microscope.

How to Observe with a Microscope.

The art of observation is at all times difficult, but is especially so with a microscope, which presents us with forms and structures concerning which we had no previous idea. Rigid and exact investigation, therefore, should be methodically cultivated from the first, in order to avoid those errors into which the tyro, when using a microscope, is particularly liable to fall. Thus, he should carefully examine the physical properties of the particles and ultimate structures he may see, and not hastily conclude that he has under observation so-called pus, tubercle, or cancer-corpuscles, because they were obtained from what was, *à priori*, believed to be pus, tubercle, or cancer. Nothing has been more clearly demonstrated by the progress of histology than the fact, that the naked sight has confounded different structures together, from a similarity of external appearance, and that the greatest caution is required at all times, but especially by learners, in forming opinions as to the nature of different tissues.

The physical characters which distinguish microscopic objects consist of—1st, Shape ; 2d, Colour ; 3d, Edge or border ; 4th, Size ; 5th, Transparency ; 6th, Surface ; 7th, Contents ; and 8th, Effects of re-agents. These we may notice in succession.

1. *Shape.*—Accurate observation of the shape of bodies is very necessary, as many of these are distinguished by this physical property. Thus the human blood globules, presenting a biconcave round disk, are in this respect different from the oval corpuscles of the camelidæ, of birds, reptiles, and fishes. The distinction between circular and globular is very necessary to be attended to. Human blood corpuscles are circular and flat, but they become globular on the addition of water. Minute structures seen under the microscope may also be likened to the shape of well-known objects, such as that of a pear, balloon, kidney, heart, &c. &c.

2. *Colour.*—The colour of structures varies greatly, and often differs under the microscope, from what was previously conceived regarding them. Thus the coloured corpuscles of the blood, though commonly called red, are in point of fact yellow. Many objects present different colours, according to the mode of illumination ; that is, as the light is reflected from, or transmitted through, their substance, as in the case of certain scales of insects, feathers of birds, &c. Colour is often produced, modified, or lost, by re-agents, as when iodine comes in contact with starch corpuscles, when nitric acid is added to the granules of chlorophyle, or chlorine water affects the pigment cells of the choroid, and so on.

3. *Edge or border.*—The edge or border may present peculiarities which are worthy of notice. Thus, it may be dark and abrupt on the field of the microscope, or so fine as to be scarcely visible. It may be smooth, irregular, serrated, beaded, &c. &c.

4. *Size.*—The size of the minute bodies, fibres, or tubes, which are found in the various textures of animals, can only be determined with exactitude by actual measurement, in the manner formerly described. It will be observed for the most part, that these minute structures vary in diameter, so that when their medium size cannot be determined, the variations in size from the smaller to the larger should be stated. Human blood globules in a state of health have a pretty general medium size, and these may consequently be taken as a standard with advantage, and bodies may be described as being two, three, or more times larger than this structure.

5. *Transparency.*—This visible property varies greatly in the ultimate elements of numerous textures. Some corpuscles are quite diaphanous, others are more or less opaque. The opacity

may depend upon corrugation or irregularities on the external surface, or upon contents of different kinds. Some bodies are so opaque as to prevent the transmission of the rays of light, when they look black by transmitted light, although they be white, seen by reflected light. Others, such as fatty particles and oil globules, refract the rays of light strongly, and present a peculiar luminous appearance.

6. *Surface.*—Many textures, especially laminated ones, present a different structure on the surface from that which exists below. If, then, in the demonstration, these have not been separated, the focal point must be changed by means of the fine adjustment. In this way the capillaries in the web of the frog's foot may be seen to be covered with an epidermic layer, and the cuticle of certain minute fungi or infusoria to possess peculiar markings. Not unfrequently the fracture of such structures enables us, on examining the broken edge, to distinguish the difference in structure between the surface and the deeper layers of the tissue under examination.

7. *Contents.*—The contents of those structures, which consist of envelopes, as cells, or of various kinds of tubes, are very important. These may consist of included cells or nuclei, granules of different kinds, pigment matter, or crystals. Occasionally their contents present definite moving currents, as in the cells of some vegetables, or trembling rotatory molecular movements, as in the ordinary globules of saliva in the mouth.

8. *Effects of re-agents.*—These are most important in determining the structure and chemical composition of numerous tissues. Indeed, in the same manner that the anatomist with his knife separates the various layers of a texture he is examining, so the histologist, by the use of reagents, determines the exact nature and composition of the minute bodies that fall under his inspection. Thus, *water* generally causes cell formations to swell out from endosmosis; whilst syrup, gum water and concentrated saline solutions, cause them to collapse from exosmosis. *Acetic acid* possesses the valuable property of dissolving coagulated albumin, and, in consequence, renders the whole class of albuminous tissues more transparent. Thus, it operates on cell walls, causing them either to dissolve or become so thin as to display their contents more clearly. *Æther,*

on the other hand, and the alkalies, operate on the fatty compounds, causing their solution and disappearance. The *mineral acids* dissolve most of the mineral constituents that are met with, so that in this way we are enabled to tell, with tolerable certainty, at all events the group of chemical compounds to which any particular structure may be referred. Other reagents are occasionally useful, such as tannic and osmic acids, magenta, glycerine, solution of nitrate of silver, &c.

Mode of Conducting the Course.

I first commenced teaching practical histology in the year 1841, and have continued doing so uninterruptedly ever since. This long experience has satisfied me that the best method of teaching the subject is to place a microscope in the hands of each student who sits at a table opposite a good light. The tables should be arranged before an elevated chair, from which the teacher may watch the manipulations of every student. Two or more experienced assistants are always necessary.

Optical illusions.—The first lesson to be acquired is how to recognise the influence of transmitted light upon transparent solid, and hollow bodies, especially in their globular, flattened, filamentous, or tubular forms. Also the influence of direct and oblique light, the use of the diaphragm, modes of illumination, and the variations resulting from the use of low, medium, and high powers. It is from a neglect of this absolutely necessary practical knowledge that we are indebted to so many errors among microscopical observers, some describing as nuclei what are only the transparent centres of homogeneous bodies, and others confounding tubes with solid fibres.

Practical investigations.—The student having learnt the optical principles on which a microscope is constructed ; the use of its various parts ; how to observe ; how to measure the magnifying power of his instrument, and of various objects ; and the optical illusions so commonly presented to his eye, is now prepared to commence his histological inquiries.

The best object to examine first is the human coloured blood corpuscle, and it is of consequence that this should be done thoroughly, and all the physical facts regarding it, including the effects of reagents, carefully described by each student. It should be also accurately measured, as I adopt its size as a standard of comparison for other structures. This accom-

plished, all the elementary, molecular, cellular, fibrous, and tubular stuctures, are similarly examined *seriatim* in the order in which they are noticed in the first part of this work, each student making and describing his own demonstrations, and the errors he may fall into at once corrected by the professor or his assistants. Then all the special organs and products of nutrition, innervation, and reproduction, are also investigated in the order in which they are described in the second and third parts of this work. As the course proceeds, the student is instructed and practised in the method of demonstrating each tissue, how to make sections, how to inject, how to prepare, stain, and put up preparations, &c. The whole subject is further illustrated by the examination of selected specimens, from a histological collection containing upwards of 3000 preparations of the vegetable and animal structures, human and comparative, healthy and morbid. These are arranged so as to form a series of demonstrations in another room, which may be examined at leisure.

Preparation of the Tissues.

Thin sections of a tissue may be made in one of three ways: (*a.*) By an ordinary *razor*, or double-edged *scalpel*. (*b.*) By a *Valentine's knife* (Plate XXI. fig. 12). This knife consists of two thin, sharp blades, *a* and *b*. They are kept opposite each other by a sliding catch, *d*, and the distance between them can be carefully graduated by means of the fine screw *c*, which passes through the blade *a*, and acts by its point on *b*. The thin section is received between the blades, and is removed by separating them at the joint *e*, and agitating them in water. (*c.*) By means of an apparatus shewn in Plate XXI. fig. 11, A. B., termed *Stirling's section cutter*.* It consists of a wooden box, *d*, having on its surface a brass plate, A (upper surface shewn at B). The interior of the box is circular, and communicates with a short tube, *b*, at the bottom of which there is a fine screw, *c*. The tissue is placed in the tube, surrounded by carrot or orange peel. The tissue may now be elevated to any extent required by turning the screw; a razor or amputating knife is placed on the brass plate and pushed obliquely through the tissue projecting above its surface. The fineness of the screw enables us to make very thin and regular sections.

* Journal of Anatomy and Physiology, May 1871.

It is often difficult to cut sections of certain tissues, owing to their brittleness or extreme softness. To accomplish this, the tissue may be surrounded by two bits of carrot, orange peel, elder pith, or spermaceti. The following paraffine mixture is useful for this purpose : Solid paraffine, 5 parts ; spermaceti, 2 parts ; axunge, 1 part. This mixture may be melted and poured into a mould, in the centre of which is imbedded the tissue under examination.

All tissues should, as far as possible, be examined in their natural condition, with no reagent except serum or aqueous humour. But it is frequently necessary to act on the tissues with chemical or physical reagents, so as to explore their structure. These reagents may be classified into (1.) physical reagents, such as heat, cold, and electricity ; (2.) chemical reagents, such as acetic acid ; (3.) hardening reagents, such as alcohol, chromic acid, &c. ; and (4.) softening reagents, such as nitric acid, maceration, &c.

1. *Physical reagents.* *a. Heat.*—It is evident that certain delicate tissues or fluids, such as nerve or blood, are best seen under conditions simulating as far as possible those in which they exist in the living body. Certain of these conditions, such as a degree of moisture and the temperature of the body, may be readily obtained by the use of an apparatus termed *Stricker's hot stage.* This consists of a strong flat brass box, accurately adapted to the stage of an Oberhauser or Hartnach microscope, having in the centre of it a small circular chamber, the bottom of the latter being made of glass, so as to permit reflected light to pass from the mirror beneath the stage. The box has inserted into each end a narrow brass tube for the ingress and egress of fluid, and attached to it there is a small centigrade thermometer which registers the temperature of the fluid in the box. Hot water is supplied to the box by means of an India rubber tube connecting it with a boiler, which, in turn, can easily be kept full by means of a tube leading from a cistern, or from one of the stop-cocks of the laboratory. It is evident that the temperature of the water in the box will be regulated by the rapidity of the flow, because if this be slow, the water has more time to cool by radiation, whereas if it be fast, a constant supply of hot water keeps the temperature high. The flow of water through the box is therefore regulated by means of an apparatus placed on the right hand side of the stage, consisting

of an upright of brass carrying a small conical glass tube, the apex of the cone being directed upwards, and drawn to a fine point. The base of the cone is connected with an india-rubber tube leading from the brass box, and the cone can be readily raised or depressed on the support, and fixed in any position by a screw. By means of this arrangement, the water is prevented from flowing rapidly through the box, and the flow can be regulated by raising or depressing the glass cone. This hot stage has been found useful in the examination of white blood corpuscles. A small drop of blood is placed on a covering glass, and a drop of tepid water having been previously introduced into the bottom of the circular chamber, the covering glass is placed over the chamber so as to form its roof, the drop of blood being on its under surface. Thus the blood corpuscles are seen free from pressure, in an atmosphere saturated with moisture, and at a temperature corresponding to the temperature of the body. The temperature may also be varied by regulating the flow of fluid through the box. The actions of various gases, such as carbonic acid or oxygen on the blood corpuscles, may also be observed by means of this apparatus. Two short brass tubes communicate with the circular chamber. One tube is for the entrance, and the other for the exit of the gas.

β. *Cold.*—A low temperature kills a living tissue, and soon hardens it. Tissues may be readily hardened by freezing. Those most suitable are lung, liver, spleen, kidney, and blood glands. The best freezing mixtures are the following : (1.) Ice. (2.) Ice, 2 parts ; chloride of sodium, 1 part. (3.) Calcium chloride, 3 parts ; snow, 2 parts. (4.) Potassium nitrate, 4 parts ; hydrochlorate of ammonia, 100 parts ; water, 200 parts.

γ. *Electricity.*—The action of either a continuous or interrupted current on a microscopical object, may be observed by the following arrangement : Two small triangular pieces of tin foil are cemented on the surface of an ordinary glass slide, of such a size that the bases correspond to the ends of the slide, and the apices approach within the 1-10th of an inch from each other. These serve as electrodes, and they may readily be brought into connection with a battery by means of wires kept in apposition to them by small, but strong, wooden clips. By this arrangement, the action of electrical currents and shocks on cilia, white blood corpuscles, or muscle, may be easily studied.

2. *Chemical reagents.*—α. The solution of *acetic acid* most serviceable, is prepared by mixing equal parts of ordinary acetic acid and distilled water. β. *Soda solution*, is caustic soda, 1 part; water, 25 parts. γ. *Glycerine* and *glacial acetic acid:* glycerine, 1 ounce; acid, 5 drops. δ. Price's pure glycerine. These all more or less dissolve and render transparent albuminous tissues. ε. *Tannic acid*, 2 grains to 1 oz. of distilled water, has a peculiar effect on the coloured blood corpuscle (p. 63).

3. *Hardening reagents.*—α. *Alcohol.*—The tissue should first be allowed to remain for several hours in dilute alcohol, and then be immersed in absolute alcohol. The retina may be hardened by changing the absolute alcohol every four hours for three days. β. *Chromic acid.*—For most tissues, a solution containing ·25 per cent. is suitable. For hardening nerve substance, a solution of chromic acid, 1 part; potassium bichromate, 2 parts; water, 1200 parts, for a week, and a fluid of twice the strength for six weeks are required (Lockhart Clarke). γ. *Müller's fluid.*—Potassium bichromate, $2\frac{1}{2}$ parts: sodium sulphate, 1 part; distilled water, 100 parts, is useful for retina, &c., the tissue being allowed to steep for four weeks. δ. *Tetroxide of osmium*, OsO_4, in solution of from 1-10th to 2 per cent. has been found to harden retina, organs of Corti, epithelium, &c. In addition to these reagents, tissues may be hardened by drying (tendon), freezing (lung), or boiling (crystalline lens).

4. *Softening reagents.*—α. *Nitric acid.*—This acid dilute (1-4 water) readily softens muscle, causing it to split up into fibrillæ or discs. β. *Hydrochloric acid* (1-20 of water) is useful for extracting the earthy matter of bone. γ. *Nitric with chromic acid.*—Nitric, 2 parts; chromic, 1 part; water, 100 parts, also soften bone or tooth. δ. *Glycerine*, with the addition of a few drops of glacial acetic acid, macerates nerve and other tissues.

Staining the Tissues.

The art of staining tissues has thrown much light on Histology by displaying structures which, without it, shew almost no trace of organisation. This depends on the affinity which certain parts of the tissues, and certain tissues, have for colouring matter. The following are the chief staining solutions:

1. *Magenta.*—This is used in the form of an alcoholic solution,

and is readily obtained by employing the fluid sold in small bottles by chemists, either dilute or not.

2. *Ammoniacal solution of carmine.*—Carmine, 10 grains; strong ammonia, 30 minims; glycerine, 2 ounces; distilled water, 2 ounces; rectified spirit, ½ ounce. Place the carmine in a test tube, add the ammonia, boil for a few seconds, let it stand for an hour, add the water, filter, and add the spirit and the glycerine. After steeping, remove the superfluous pigment by allowing the tissue to be immersed in an aqueous solution of glycerine, 2 parts of glycerine to 1 part of water. The pigment may be fixed by placing the tissue in acid glycerine (glycerine, 1 ounce; hydrochloric acid, 2 drops; glacial acetic acid, 5 drops).

Logwood. — Pulverise the ordinary extract, and add three times its bulk of alum. Mix well for twenty minutes in a mortar, first with a small, and then with a larger quantity of water. This, when filtered, should present a clear, dark violet colour. If it be a dirty red, add more alum. To one ounce of this fluid two drachms of 75 per cent. alcohol must be added. A few minutes will stain tissues with this solution even when previously hardened by alcohol or chromic acid.

3. *Nitrate of silver.*—A solution of ½ per cent. in distilled water is used for demonstrating the margins of epithelial cells in lymphatics, blood vessels, or on peritoneum. The tissue is steeped in it for two or three minutes, then in very dilute acetic acid (1 to 2 per cent.) for a minute or two longer, then placed in glycerine and exposed to the action of the light.

4. *Chloride of gold*, ½ per cent. solution, stains nerve tubes. The tissue is steeped in it for 20 minutes, and then washed in dilute acetic acid (1 to 2 per cent.).

In the manipulation of tissues by these various agents, it must be remembered that we frequently change their natural appearances. It would be as absurd to suppose that a nerve tube, with its solid central rod formed by coagulating fluids, exists in the living body, as it would be to suppose that a hard-boiled egg resembles the developing ovum.

Injection of the Tissues.

The art of making successful injections can only be acquired by long practice, and by attention to many minute details, which must be taught during the performance of the operation. For this reason, only the compositions of a few of the principal

injecting fluids will be given, and some general observations made. Injections may be either *opaque* or *transparent*, the former being employed when the tissue is thin and delicate; the latter, when the form of the tissue or inequalities on its surface are to be displayed. The fluid may be driven into the vessels either by a brass syringe made for the purpose, or still more slowly and equally by hydrostatic pressure. The former method is the one most generally adopted, and by it, with a skilful hand, good results may be obtained: the latter has been recently revived by an elaborate apparatus used by Ludwig, in which the hydrostatic pressure derived from the water-pipes of the laboratory may be measured by means of mercurial manometers. Injections should be made while the body is fresh, after the disappearance of *rigor mortis*. The chief points to be attended to are: 1. That the animal, or organ, and the injection, nozzles, and syringe, should be kept warm, at a temperature sufficient to guarantee a free and uniform flow of the fluid; and 2. that the pressure made should be moderate in amount, and slow and equable.

1. *Opaque injections.*—The liquid or menstruum is composed of melted wax or melted size. Ordinary sealing wax, diluted somewhat with turpentine, or, better still, with oil of rosemary, may be employed. If pure white wax be used, the greatest pains must be taken in grinding and mixing the colours. Size is prepared by melting 4 oz. of the best transparent glue in one pint of water, and then adding to it the colour, which must be kept constantly stirred. The advantage of wax injections is, that when cold, the vessels remain fully distended and round, while sections of the tissues may be kept hard. The finest preparations of this kind have been made by the injectors of the Vienna school, ever since the days of Lieberkühn; and the present eminent professor of that university, Hyrtl, continues to make the most beautiful injections in wax, after the method of his predecessors. Size injections, on the other hand, shrink when allowed to get dry, and hence they must always be kept moist in bottles, or, for microscopic purposes, mounted in closed cells with diluted spirit. The preparations made by Quekett and Hett in this manner have never been surpassed in beauty. The various colours employed are as follow:—

a. Vermillion.—This should be carefully ground down on a slab with oil or water, and all the coarser particles removed by

lixiviation. To secure success, the ground paint should be examined microscopically, for if the particles are not much smaller than the blood corpuscles, they will not pass through the capillary vessels. 2 oz. of vermillion are to be added to 1 pint of the liquid or menstruum, and kept constantly stirred till used.

b. Chromate of lead.—This is obtained by dissolving 200 grains of acetate of lead and 105 grains of bichromate of potash in equal quantities of water, mix, pour off the supernatant fluid (acetate of potash), and mix the chromate of lead with 4 ounces of hot size. M. Doyère's method is to throw in saturated solutions of the two salts, one after the other, so that the formation of chromate of lead takes place in the vessels. It has been found, however, better to add gelatine in the proportion of 4 ounces in 8 ounces of water, to 8 ounces of the saturated solutions of each salt.

2. *Transparent injections.*—The fluid or menstruum employed here is a size made of transparent gelatine or of the compositions given below.

a. Beale's Prussian blue fluid, consists of the following parts: glycerine, 1 ounce; spirits of wine, 1 ounce; ferrocyanide of potassium, 12 grains; tincture of perchloride of iron, 1 drachm; hydrochloric acid, 5 drops; water, 4 ounces. Dissolve the ferrocyanide of potassium in 1 ounce of water, and the tincture of the perchloride of iron in another. Mix these gradually in a bottle, adding the iron to the ferrocyanide of potassium. Next the spirit, the glycerine, and the rest of the water, are to be mixed, and the colourless fluid thus obtained is to be well shaken up with the Prussian blue.

b. Turnbull's blue.—Ferricyanide of potassium, 10 grains; sulphate of iron, 5 grains; water, 1 ounce; Price's glycerine, 2 ounces; alcohol, 1 drachm. The iron is dissolved in the glycerine and water, and the ferricyanide of potassium and alcohol added.

c. Beale's carmine fluid.—Carmine, 5 grains; glycerine (with 8 or 10 drops of hydrochloric acid), ½ ounce; glycerine, 1 ounce; alcohol, 1 drachm; water, 6 drachms; ammonia, a few drops. Mix the carmine with a little water, and add 5 drops of *liquor ammoniæ.* To this add ½ oz. of glycerine, and shake. Add gradually the acid glycerine. Add the alcohol and water gradually, shaking the bottle thoroughly.

d. Carter's carmine fluid.—Pure carmine, 60 grains; liquor

ammoniæ fortior, 120 grains; glacial acetic acid, 86 minims; solution of gelatin (1-6 of water), 2 ounces; water, 1½ ounces. Dissolve the carmine in the ammonia, and filter. Mix this with 1½ ounces of hot solution of gelatin. Mix the remaining ½ ounce of gelatin with the acetic acid, and drop it slowly into the solution of carmine.

e. Thiersch's yellow fluid.—Dissolve 10 parts of bichromate of potash in 110 parts of water, and make a solution of nitrate of lead of the same strength. Mix 1 part of the solution of bichromate of potash with 4 parts of a concentrated solution of gelatin, and 2 parts of the solution of nitrate of lead with 4 parts of gelatin. Mix these two gelatin solutions at a temperature of 75° to 90° F., and afterwards heat the mixture for half an hour to 212°. Then filter.

Preservation of the Tissues.

Many preservative solutions have been used, and different tissues require different fluids.

1. *Canada balsam.*—This substance has been more used than any other. It is best prepared by drying pure Canada balsam in a porcelain capsule in a hot-air chamber, until it becomes quite hard, and then dissolving the mass in chloroform or turpentine.

2. *Dammar fluid.*—Gum Dammar, ½ ounce; oil of turpentine, 1 ounce; dissolve and filter. This is then mixed with the following: gum mastic, ½ ounce; chloroform, 2 ounces; dissolve and filter.

3. *Potassium acetate.*—A saturated solution of this salt is the best medium for preserving all tissues which have been acted on by osmic acid.

4. *Glycerine.*—This substance, either natural or containing 1 or 2 drops of hydrochloric acid to the ounce, is suitable for preserving tissues not made too transparent by it.

5. *Glycerine jelly.*—Allow a certain quantity of gelatin to soak for 10 hours in cold water, when it will be found swollen and soft. Melt in warm water. Add an equal amount of strong glycerine. This is suitable for cartilage, blood vessels, kidney, &c.

6. *Weak spirit* (1 part spirit, 3 parts of distilled water) is well suited for muscle, blood vessels, &c.

7. *Naphtha and creosote.*—Creosote, 3 drachms; wood naphtha, 6 ounces; distilled water, 64 ounces; chalk, a sufficient quantity.

Mix the naphtha and creosote together, and add as much chalk as will make a paste. Add water and mix thoroughly. Add a few bits of camphor, the size of a hazel-nut, and allow the whole to stand for two or three weeks. Pour off the supernatant fluid for use.

These are the principal fluids in which microscopical preparations may be preserved. Cells for mounting and preserving microscopic preparations may be made with Brunswick black, Canada balsam, Dammar varnish, or glass. Those made of Brunswick black are objectionable, because in course of time the material runs into the cell, and the preparation is spoiled. Canada balsam sometimes cracks in old preparations, and allows the preservative fluid to escape ; at other times, it softens, and the covering glass is easily displaced. On the whole, cells made of thin glass are the best for all purposes.

These, and numerous other practical details, it is useless to describe in words, as they can only be learned in the laboratory.

PRACTICAL EXPERIMENTAL PHYSIOLOGY.

This department of Physiology includes a description of the methods of research followed and the instruments employed in the investigation of physiological phenomena. Of late years it has advanced with great rapidity, and has been fruitful in many important discoveries. When taught practically, the student should, as far as possible, assist personally in the experiments, acquire a knowledge of the apparatus employed, and learn how to observe and record results.

Experiments on the Muscular System.

As we have seen (pp. 80, 82), living muscle possesses two properties—contractility and electro-motive power. The investigation of these functions requires the use of electrical and other apparatus, which we shall now describe. To understand them, it is necessary that the student should, in the first instance, become acquainted with the general phenomena of magnetism and electricity, as already explained at pp. 144 and 146.

Apparatus.

1. *Batteries.*—These have already been described at p. 152. Daniel's battery is useful where we wish to have a current con-

stant in quantity. Bunsen's is too powerful for most physiological experiments. Grove's battery is powerful and constant. It is desirable to have eight or ten small elements of this description for experiments in which we wish to graduate the strength of the current. Smee's battery is much used, because it requires only one fluid, and is easily manipulated, but it has the disadvantage of being inconstant; so that in comparative experiments it ought not to be employed. It is important in using any of these batteries to remember that the zinc requires frequently to be amalgamated, a process easily performed by rubbing over the zinc with a little flannel or cotton wool dipped in mercury and dilute sulphuric acid. If this be not done, the zinc, which is usually contaminated with other metals, will not be homogeneous, and numerous small galvanic circles will be formed between these impurities and the particles of zinc, leading to rapid wasting of the latter. Where very careful amalgamation is required, the following fluid is useful: *Berjot's amalgamating liquid.*— Dissolve at a gentle heat, 200 grammes of mercury in 1000 grammes of a mixture of 1 part by weight of nitric acid and 3 parts of hydrochloric acid, and then add 1000 grammes of the latter.

2. *Du Bois-Reymond's electromotor or induction apparatus.*— The production of electricity by induction has been described at p. 155. A side view of the instrument is seen in Pl. XIX. fig. 1, and an end view in Fig. 2. It consists of a primary coil, R_1, of thick copper wire, insulated with silk, and of a secondary coil, R_2, consisting of numerous coils of fine copper wire, also well insulated. The centre of the primary coil, R_1, contains a bundle of thin iron wires, which are rendered magnetic during the passage of the electrical current round the primary coil, and thus increase the amount and intensity of the induced current obtained from the secondary coil (Fig. 2 S). The primary coil is firmly fixed; but the secondary one slides in a double groove in the board B B, and this board has a hinge which allows one-half to lie under the other, as depicted in Fig. 1. When, however, we wish to increase to a greater extent the distance between the primary and secondary coils, we unloose a hook, seen on the board under R_1, and fold out the board to double the original length. A scale, divided into centimetres and millimetres, is pasted on one side of the upper surface of the board, and we can thus, in comparative experiments, carefully graduate the distance between the two coils. The nearer the coils

are to each other the more intense the shock, and *vice versa.* The shock of induced electricity can, however, only be obtained at the moment of opening and at the moment of closing the primary current. This regular opening and closing of the primary circuit is effected by an apparatus placed at the end of the instrument, termed *Wagner and Neef's hammer.* This apparatus will be understood by referring to Plate XIX. fig. 2. Here we have K representing the battery from which voltaic electricity is derived. It passes from the positive pole in the direction of the small arrow to the brass pillar *a.* Having run up this pillar, it goes along a steel spring seen over S, and as the elasticity of the spring keeps a small square bit of platinum on its upper surface in apposition to the platinum point of the screw S_1 the current passes into S_1 from thence by a copper wire to S_{11}, and from thence, as indicated by the small arrow ——>, round the wire of the primary coil, R_1. It now goes from R_1, as indicated by the lower small arrow, to a small U-shaped piece of soft iron surrounded by the wire, and thus converts it into an electro-magnet, which draws down the armature of soft iron (the hammer) on the free end of the spring, and thus the contact between the back of the latter and the screw point S_1 is broken. When this happens, the primary current is broken at S_1, the electro-magnet loses its magnetism, releases the spring, which flies up by its elasticity, and again establishes the current. Thus by the alternate breaking and forming of the primary current, a secondary current is induced in R_2, which has no connection by wire with R_1. It is the secondary current (Faradic electricity) which is used in many physiological experiments.

Helmholtz's modification of the apparatus.—When a muscle or nerve is irritated by an induced current from the secondary coil, R_2, it receives a rapid series of shocks, because the hammer acts with great rapidity. If the hammer acted slowly, so that we could distinguish between the effect of the opening shock and the closing shock, it would be found that the effect is much greater in the case of the former. The reason of this is that the opening shock is more rapid in its course, its velocity rising rapidly from zero to a maximum. If then we can retard the opening shock, we may render it equal to the closing one. This Helmholtz has accomplished by an arrangement in which he makes the hammer, when attracted by the electro-magnet,

not open the primary circuit, as in the apparatus just described, but close an accessory circuit so as to weaken the primary one. The accessory circuit (Pl. XIX. fig. 2) is β running from a to S_{III}. Beneath the centre of the steel spring is a middle pillar, having in its base the binding screw x to which a wire in connection with the negative pole of the battery is attached. The accessory circuit, $a\ \beta\ S_{III}\ S_{II}\ R_1$, continued in the direction of the lower arrow down an electro-magnetic pillar to another arrow which leads to x, S, and a, is closed when the steel spring is in contact with S. When this is the case, the primary circuit is so weakened that the electro-magnet loses its magnetism, and the spring flies up, the primary circuit being now formed and the accessory broken. By this arrangement the velocities of the two shocks are equal, and the apparatus is better adapted for careful physiological research.

3. *Du Bois-Reymond's key* (Plate XX. fig. 4).—When we wish to open or close a circuit, we may do so either by detaching or fixing the wires by binding screws, or by using small cups filled with clean mercury, into which the ends of the wires dip. This latter expedient is adopted in very delicate stimulation experiments. It is, however, preferable in most experiments to use the galvanic key. It consists of a plate of vulcanite firmly attached to a strong rectangular vice-pin. On the plate there are two small thick pieces of brass, each bearing two binding screws, *b*, *c*, placed at a short distance from each other. These can be united by raising or depressing the brass arm, *d*, which has an ivory handle, and turns on a pivot attached to the piece of brass, *c*. The wires from the battery are connected with the two internal binding screws, while the two external ones connect those going to the nerve or muscle. When the handle is pushed back, the key is opened, and the current passes to the tissue to be irritated; but when the handle is pushed forwards and the key closed, as seen in the figure, the current passes along the thick arm of brass, back again to the battery.

4. *Pohl's commutator, gyrotrope, or rheotrope* (Plate XX. fig. 6).—This instrument is for the purpose of inverting or changing the direction of a current of electricity. It consists of a round disc of wood or vulcanite having six small mercury cups, A, B, a, b, α, β, in it substance, in connection with each of which there is a binding screw. The cups a, b, are in con-

stant connection with a pair of brass wires, P, O, brought close to each other at S by a piece of glass tubing, into which they are inserted. These brass wires carry two brass arcs transversely, *m*, *n*, *o*, and *p*, *q*, *r*, having their ends free, so that by moving the glass bridge S forwards or backwards, the ends may dip into the cups α, β, or A, B. The apparatus may be used with or without the cross wires *h*, *i*, the wire *h* being curved in the middle so as not to touch *i*. (1.) *Without the cross wires.*—The wires from the battery or key are fixed into *a* and *b*. The current cannot pass across the bridge S, because the centre is composed of glass. If now two wires are in connection with α, β, and the arcs dip into these cups, as shewn in the figure, the current will flow along those wires. If, on the other hand, two other wires are in connection with the cups A, B, and the ends of the arcs are dipped into them, then the current will pass from A to B. Thus, by simply *turning* the central bridge bearing the arcs, we can send a current either in the direction of α, β, or A, B, at pleasure. (2.) *With the cross wires.*—Suppose, now, the wires *h*, *i*, are inserted so that the wire *i* connects the cups α, B, and the wire *h*, β and A, a different use is made of the instrument. If the current, for example, entered at $+a$, it would follow the direction *a*, P, *r*, α, β, *n*, *o*, and back to the battery by—*b*. If now we reverse the bridge, the direction will be *a*, P, *q*, A, along the cross bar *h* to β, α, from thence along the cross bar *i* to *m*, *o* and $-b$, and from thence back to the battery. By this last arrangement, the current has passed along the circuit between α and β in such directions that it can be sent up or down a nerve at the will of the experimenter. The course of the current, however, in these cases, can only be clearly understood by a study of the instrument itself.

5. *Muscle telegraph* (Plate XIX. fig 7).—This is an apparatus very useful in experiments on both muscle and nerve, and it consists of the following parts: A brass forceps, A, at the end of an arm capable of sliding up and down on an upright round pillar of brass. This is for holding the femur of a frog's leg which has the gastrocnemius muscle attached to it. The screw S, permits the forceps A, to be rotated in any direction, and fixed securely in any position. Into the tendon of the gastrocnemius a small hook *h* is inserted, to which is attached a fine thread passing round a pulley *p*, and then downwards along *a* to a little balancing bucket *b*, containing a few shot. On the

same pulley we have a long arm, bearing at its free end a round coloured disc of mica, which moves in front of a piece of white plate in the direction of the arrow. By the contraction of the muscle, the signal is pulled up to a greater or less extent, and thus the effect of irritation on it may be seen by many observers at once. If the muscle is to be stimulated by the direct application of a current, the wire from the positive pole is *a* attached by the bending screw S, and that in connection with the negative, *x*, is wound tightly round the hook fixed in the *tendo Achillis*. The upright bearing the pulley and disc, is fixed in a wooden socket which can move in either direction in a groove, and is retained in its position by a strong screw, Z.

6. *Du Bois-Reymond's polarizable electrodes* (Pl. XIX. fig. 6).—This apparatus is designed for the purpose of stimulating a nerve in any situation. It consists of *a*, a strong wooden stand bearing a round piece of vulcanite *b*, in which we have two small binding screws. In connection with this we have a long arm, having in its centre a universal joint which can be tightened in any position by the screw *c*. This bears the essential part of the apparatus, which consists of two triangular platinum points or plates, *e*, each soldered to a thick wire passing through a square block of ivory. This block has two small screws on its upper surface, which are for the purpose of adjusting the distance of the platinum points from each other. Underneath the ivory block and electrodes we have a glass plate for insulating the wires. When the instrument is used, the nerve is laid across the two electrodes, and the portion of nerve on and between them is thus stimulated.

Experiments on Contractility.

The property of contractility can be readily demonstrated by shewing that a piece of living muscle, or a whole muscle, contracts when irritated. The irritation may be mechanical, such as by a blow, or a pinch; chemical, as by the action of a solution of common salt; electrical, on applying electricity; or vital, on stimulating a nerve. The truth of the Hallerian doctrine of contractility, namely, that it is inherent in muscular tissue, and not dependent on nerve may be demonstrated by two experiments.

1. *John Reid's experiment.*—Remove the sciatic nerve from the leg of a living frog, and irritate the muscles by an induced cur-

rent of electricity. At first there is violent spasm or tetanus, but after some time the muscles cease to respond to the stimulus. Thus the contractility has been exhausted. But allow the frog to live for six, eight, or twelve hours, and it will be found, on again applying the stimulus, that the contractility has returned, while sensibility has not.

2. *Bernard's Woorara experiment, as shewn by Du Bois-Reymond.**—This requires the following apparatus : 2 muscle telegraphs, 1 of Du Bois-Reymond's polarisable electrodes, a Pohl's commutator, without the transverse bars, a key, an induction apparatus, and a Smee's or Daniel's element.

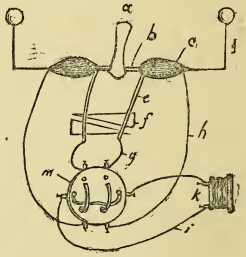
Mode of preparing the frog.—The frog is chosen for many of these experiments because it is easily manipulated, and the muscles and nerves can be quickly isolated. First, decapitate the animal, or cut through the *medulla* with a sharp pair of scissors, to destroy sensation, then holding it in the left hand by the legs or lower part of the body, turn the animal round, and cut the body through about the middle of the abdomen. Then sieze hold of the back bone with the fingers and thumb of the left hand, and with the forefinger and thumb of the right hand, quickly drag off the skin of the legs. Then various preparations may be made according to the nature of the experiment. The limbs may be used without further dissection, or the sciatic nerve may be dissected out (Plate XIX. fig. 3 *b*), or the gastrocnemius muscle, with or without the sciatic nerve, may be isolated. In the experiment about to be described the sciatic nerve is first dissected out. This is easily done by pressing the muscles of the thigh forwards with the thumb and forefinger of the left hand, when the nerve usually starts into view. Then with a pair of blunt-pointed scissors, separate the nerve from the surrounding tissues, taking great care not to injure or even touch it. Cut through a small branch which runs downwards and inwards a little above the middle of the thigh. Trace the nerve as high up as possible, so as to have it of considerable length, cut it, and with a blunt quill or an ivory point turn it down upon the gastrocnemius in order to keep it moist. Then remove the muscles of the thigh, except the attachment of the gastrocnemius, and snip through the femur just below its head. Now cut through the *tendo Achillis*, and

* Leçons sur les effets des substances toriques, 1857, p. 277.

pull the gastrocnemius upwards to the knee. Then amputate the leg below the knee, and you have a preparation consisting of a femur having the gastrocnemius attached to its lower end, together with the uninjured sciatic nerve. A hole in the gastrocnemius tendon should now be made for the steel hook of the muscle telegraph.

Woorara solution.—Dissolve 5 grains of woorara in a little weak spirit, rubbing it up in a small mortar, and then add 5 drops of glycerine and three drachms of distilled water. Each minim contains about the 1-38th part of a grain, and usually 6 minims, or about the 1-6th part of a grain is a sufficient dose for the experiment.

The experiment consists in first putting a ligature on the femoral artery in one of the limbs of a frog, or tying a tight cord round the upper part of the limb, so as to prevent the poison entering it; then with a syringe having a fine nozzle we inject, under the skin of the back, six minims of the above solution, and allow the animal to come thoroughly under its influence. It should take about half-an-hour to completely prostrate the animal, so that it rests on its belly unable to move. Care must also be taken not to inject too strong a dose of the woorara. Two dissections are now made in the manner already described. In the one the sciatic nerve has been poisoned by woorara, and in the other, it is quite healthy. Each limb so prepared is now attached to a muscle telegraph (see Fig. *d*), a pair of brass forceps being placed midway between the two telegraphs, so as to hold the two femurs (*a*), and the two nerves (*e*) are laid across the platinum points of Du Bois-Reymond's electrodes (*f*). These electrodes are connected by wires (*g*) with two of the cups of Pohl's commutator by binding screws, and from the other two cups, wires (*h*) proceed to, and are wound round, the hook fixed into the *tendo Achillis* of each muscle (*c*). The two cups of the commutator, into which the wires bearing the arcs are permanently fixed are connected with two outer binding screws of a key placed at *i*. From the two



inner binding screws, wires (*i*) proceed to the secondary coil of an induction apparatus (*k*), while the primary coil is placed in the circuit of a Daniel's or Smee's battery, consisting of one cell. By this arrangement of apparatus, the commutator enables us to reverse the electric current so as to irritate the two nerves or the two muscles at pleasure. If successful, the result will be that when both nerves are irritated, only one muscle contracts and elevates the disc of a telegraph, namely, the one supplied by the nerve not poisoned by woorara; whereas, if the current be sent through the *muscles*, both contract, and the two discs are elevated. Thus, although the nerve has been poisoned by woorara, the contractility of the muscle remains, a fact which demonstrates the truth of the Hallerian doctrine that it is a property inherent in muscle, capable of being excited by any direct stimulus, and not dependent on the nervous system.

Kölliker's experiment.—A reverse experiment to the one just described is made by destroying the contractility without affecting the motor nerves by means of *veratria*. The experiment is conducted in the same manner, and it will be found that when both *nerves* or both *muscles* are irritated, one muscle contracts, namely, the one which has not been poisoned.

Experiments on the evolution of electricity by muscles.

All muscles evolve a constant stream of electricity (p. 82), passing from any natural or artificial longitudinal section to any transverse section (p. 166). In order to demonstrate this important fact, the following apparatus is necessary :

(1.) *A multiplying galvanometer* (see p. 149).—This instrument is represented in Plate XIX. fig 4. It consists of an astatic combination of two magnetic needles (p. 149), the lower of which is surrounded by a coil of fine copper wire. When even a feeble current of electricity is passed through this wire, a deflection of the needle is obtained. The instrument must be carefully arranged, the needles rendered perfectly astatic so as to point from east to west, and the whole levelled by means of three screws supporting a brass disc (*a*) : on the surface and in the centre of the disc is a brass box (*b*), on the top of which is fastened a boxwood frame (C) supporting the coils of wire. This brass box can be made to revolve by means of a screw (*g*), which

acts as a fine adjustment, and enables us to place the coils of wire in any direction we please. The disc (*a*) is usually graduated into 1-10ths of a degree. The two pillars (*hh*) support a horizontal bar, having suspended from its centre, by a single fibre of spun silk, the astatic combination of needles, which are usually kept steady from vibrations by a small magnet placed on a line with the direction of the coils of wire. On the top of the wooden frame (C) we have a circular scale of white paper divided into parts of 90°, which is so arranged that zero is also parallel with the direction of the coils of wire. The instrument is protected from dust and currents of air by means of a cylindrical glass cover. It must be securely fixed, carefully levelled, and made free from vibration.

(2.) *A pair of Du Bois-Reymond's non-polarizable electrodes.* (See Plate XIX. fig. 5.)—They consist of shallow troughs of zinc, carefully amalgamated on the inner surface. In connection with each trough there is a brass pillar (*c*) supporting two binding screws. The trough is placed upon a piece of vulcanite which acts as an insulator. Into each trough we place a saturated solution of sulphate of zinc. We must now prepare two cushions of Swedish filter-paper as follows: Fold a sheet so as to make a bundle or cushion about half an inch thick. Place it in the trough so that one surface rests everywhere in close contact with its bottom, bend one side over, as shewn in Fig. 5, *b*, and with a sharp razor cut the cushion so as to make a perpendicular surface. The cushions being thoroughly saturated with the sulphate of zinc solution would exercise an irritant action on the muscle, if laid upon them, and would cause it to contract. This is avoided by making two thin films or plates of sculptor's clay moistened with saliva. One is placed on the surface and perpendicular section of each cushion, as seen in Plate XIX. fig. 5 (g). Strips of bladder well soaked in white of egg may also be used for this purpose.

(3.) *Arrangement of apparatus.*—The two troughs, prepared as already described, are placed opposite to each other, at a distance of about a quarter of an inch. A thin wire is conveyed from each trough to the two innermost binding screws of a key. From the two outer binding screws, wires pass either directly to the galvanometer or to a special apparatus termed the *commutator*. This is a mahogany box, having inside a coil of wire to increase the resistance to the electrical current, and a

series of brass plates and movable contact levers connected with each other, so that the muscular current passing through it can be diminished or inverted at will.

In order to prove that the apparatus does not itself produce any electric current, the two troughs should be connected with each other by a little oblong pad of blotting paper wet with the solution of zinc sulphate. The key is now opened, and if the apparatus be in order, the needle is unaffected. Any long muscle of a frog can be dissected out, the gastrocnemius is best, and a clean transverse section made with a pair of sharp scissors. The piece of muscle thus prepared, is now laid upon the plates of moist clay on the cushions in the troughs, so that its transverse section is placed accurately against the one cushion, and its longitudinal section against the other. The key is now opened, and a deflection of the galvanometer needle at once indicates the presence of a current of electricity, and the direction in which one pole of the needle is deflected shews the direction of the current. The key is now closed, and when the needle, after oscillating, stops at zero, the position of the piece of muscle should be reversed, and the key opened, when it will then be found that the needle will be deflected in the opposite direction to that of the first experiment. The experiment may be modified by placing a *transverse* section, or a longitudinal section, against each cushion, when there will be no or little effect on the needle. For this purpose, and for ascertaining the various electrical conditions of points of the surface of the muscle, there are many subsidiary contrivances, termed *supporting plates*, *rheophoric tubes*, &c., a knowledge of the use of which is best acquired by practice.

It having thus been clearly shewn that a piece of living muscle evolves a current of electricity, it may next be demonstrated that when the muscle is thrown into a state of contraction, this property is diminished. For that purpose it is necessary that the muscle have attached to it the uninjured sciatic nerve. It should then be adjusted on the two cushions, as previously described, while the nerve is laid gently on the platinum electrodes, which are in connection with an active induction coil, a key intervening. Deflection of the needle by the muscular current is now allowed to take place, and then to come to rest. On opening the key between the battery and the platinum electrodes, the muscle on the cushions is at once

thrown into a state of contraction through the irritation of the nerve, and the needle of the galvanometer will be seen returning towards zero. Thus it is proved that the intensity of the electro-motive power is opposed to that of active contractility. (See p. 168.)

Numerous other structures, both animal and vegetable, such as skin, intestine, stems, leaves, bark, &c., may be examined with the view of ascertaining the presence or absence of a current of electricity.

Du Bois-Reymond's experiment to shew the presence of a muscular current in the living human body.—This will be understood by referring to Plate XIX. fig. 10, where an individual is represented grasping a roller, *c*, attached to two wooden supports firmly screwed to the table, so that the forefinger of each hand is immersed in, and touches the bottom of, the trough *b*. If the muscles of both arms are relaxed, there is no effect on the needle of the galvanometer *a*, but if the muscles of one of the arms are contracted by firmly grasping the roller, in many individuals we see a feeble deflection of the needle. Among the numerous students attending the practical physiology class during each session, a very few are found capable of causing this deflection, and we have observed that a large proportion of the successful individuals are of what is usually described as the sanguine temperament, having often red hair.

Experiments on the effects of muscular irritation.

This investigation, which is of great importance therapeutically, should be conducted in the following manner :

(1.) *Effect of a continuous current* (p. 82).—The hind leg of a frog is held by a pair of strong brass forceps, sliding on a round pillar of brass which is fixed into a solid wooden stand. Such a pair of forceps may be seen on the left side of fig. 7, Plate XIX. A. The limb so fixed is to hang down loosely, the knee being flexed. Two wires from a Daniel's or Smee's element are attached to the inner binding screws of a key. Wires are also conducted from the two outer binding screws, one being fixed by the screw S (Plate XIX. fig. 7), while the other is attached to the frog's foot. It will now be found that the muscles of the limb contract on opening and on closing the key, but there is no contraction while the key is open, that is during the passage of the continuous

current. A continuous current, however, effects electrolytic changes in the muscle.

(2.) *Effect of an interrupted current.*—By the same arrangement of apparatus, it may be shewn that an interrupted current stimulates the muscle on the opening and closing of the key, and if this be done so rapidly that the muscles have not time to become relaxed before they receive a fresh shock, a constant state of rigidity or *tetanus* is occasioned.

(3.) *Effect of an induced or Faradic current.*—When an induction apparatus is employed for stimulating the muscle, the latter at once becomes tetanic, even although the same galvanic element is used to produce the induced current, as was employed in the two previous experiments. This shews that an induced current irritates much more strongly than either a continuous or an interrupted current.

Production of tetanus.—Tetanus is a state of permanent muscular contraction. It may be studied by the following illustrative experiments :

(1.) *By mechanical violence.*—Kill a frog by violently striking its head against the table. It will usually be found that for some minutes after death, the body becomes rigid from the tetanic condition of the muscles.

(2.) *By an induced current.*—As has been above described, tetanus is produced by an induced or *Faradic* current.

(3.) *By saline solutions.*—Prepare the limb of a frog in the usual way, and expose the sciatic nerve. Place the limb on a glass plate, and allow a drop of a strong saline solution to fall upon the nerve. In a few minutes the muscles become tetanic from the irritant action of the saline solution on the nerve.

(4.) By *Kuhne's apparatus*, consisting of a pair of vertical forceps fixed over a circular glass plate, the irritant action of many different acid, alkaline, and saline substances can be examined. The muscle preparation is fixed in the forceps, so that either it or the nerve are suspended over the glass plate. The latter may be elevated or depressed by a screw placed underneath it, so that the irritant solution, the effect of which is to be examined, may be brought into contact with the extremity of the muscle or nerve.

(5.) *By Heidenhain's Tetanometer.*—Another very ingenious mode of producing tetanus is effected by the use of an apparatus termed *Heidenhain's hammer* or *tetanometer*. The use of this

instrument is to beat the nerve supplying a muscle or limb with great rapidity, and thus, by frequent mechanical irritations, produce tetanus. It is shewn in Plate XIX. fig. 9, and consists essentially of a modification of Neef's hammer, already described in connection with the induction coil (p. 527). The apparatus rests upon a vulcanite plate, K, supporting a brass column, a, which carries the lever L, in the middle of which is the armature of the electro-magnet (near L). This lever turns upon an axis at a, and may be relaxed or tightened by turning the screw S, which acts upon a spiral wire above it, marked with a smaller s. The pillar C supports, on a horizontal arm, the screw S_{i}, the point of which rests on a small piece of platinum on the upper surface of the lever. It is at this point the current is broken, and thus the hammer caused to vibrate. In the base of C we have a screw, S_{iii}, which secures the wire coming from the positive pole of a galvanic element, while the screw Z receives the wire in connection with the negative pole. When used in the present experiment, the screw S_{ii} is attached, by a piece of copper wire, with the screw S_{iii}. The hammer, properly so called, is seen at *h*. It is made of ivory, and beneath it there is an ivory support, *t*, which receives the nerve in the groove h^{1}. Behind the ivory support there is a small ivory axle, A, fixed between two brass notches, and caused to move slowly by the pressure of the spring *p*. By means of the screw S''', the whole of this part of the apparatus may be elevated or depressed at pleasure. When the sciatic nerve of a limb (which is placed on a glass-support, or held by a pair of forceps before the apparatus) is placed in the ivory groove, and rapidly beaten by the ivory hammer, which is worked by the electro-magnet, the muscle becomes at once tetanic. The nerve is usually rapidly beaten through, and then it is necessary, in order to repeat the experiment, to attach its end by a fine silk thread to the ivory axle, and by turning the latter, to drag through a fresh piece of nerve. The only difficulty in using this apparatus is, that the different parts must be so adjusted with reference to each other that the nerve be beaten with a proper amount of force and rapidity.

(6.) *By Poggendorff's wheel.* — Another mode of producing tetanus by an interrupted current is by means of an instrument termed *Poggendorff's wheel.* This instrument consists of a

wooden disc or wheel having twenty or more spaces cut out of its circumference, which are occupied by pieces of brass, alternating with pieces of ivory. The pieces of brass are connected alternately with each half of the axle, the two halves being insulated from each other in the centre of the wheel by an intervening piece of vulcanite or glass. Two stiff brass springs, having screws at their bases, are caused to press firmly against the margin of the wheel, so that while the wheel is being rotated, the flattened ends of the springs touch alternately the pieces of ivory and the pieces of brass. The binding screws in connection with the springs receive wires coming from a galvanic element, while there are two others on the supports of the axle from which wires may be conducted to a muscle. When the connections have been thus made, and the wheel rotated slowly, the muscle contracts and relaxes alternately, but when turned quickly, it becomes tetanic.

(7.) *Ritter's tetanus.*—It may also be demonstrated that if a continuous current of electricity be allowed to traverse a nerve attached to a limb in the upward direction, that is centripetally, for two hours (the nerve being kept moist and warm), and the current be then suddenly interrupted by moving the electrode *next* the free end of the nerve, the muscles become suddenly tetanic. This has been termed the *tetanus of Ritter.*

(8.) *By strychnine.*—Tetanus may also be produced by poisoning a frog with a solution of strychnine. This poison, injected hypodermically, increases the reflex action of the spinal cord, so that the slightest irritation at once throws the animal into a state of tetanus.

Effects of the opening and closing induction shock on a muscle. (See p. 82.)—This may be studied accurately with the aid of an instrument termed *Pflüger's falling apparatus* or *trip-hammer.* A view of this instrument is seen in Plate XIX. fig. 8. On a plate of vulcanite, E, there is a brass plate, having two uprights, *d d*, between which we have the steel axle of the hammer *e.* This hammer consists of a long rectangular handle, *a'*, and of a large steel head, *i.* On the left side of the latter we have a small steel rod, *m*, pointing downwards, and tipped with platinum. Above the hammer head, there is an electro-magnet, A, placed between the two supports *d d*, which may be fixed at any height by the screws *n n.* When this electro-magnet is called into action by a Smee's element, the head of the hammer is

supported as seen in the figure. If, however, the galvanic circuit be opened, the hammer head falls and strikes against the end of the lever P at *q*, forcing it downwards, and thus breaking the contact of the other end of the lever with the screw-point *r*. At the same moment, the platinum point *m* falls into the conical steel trough X, which contains mercury. By this arrangement the instrument may be introduced into two galvanic circuits, and the moment the hammer head falls, the one circuit is closed and the other opened. The connections are made, for the first circuit, by wires attached by the screws *c* and *y*, and for the other by wires attached to the screws *t* and *u*. Suppose the hammer head to be in the position represented in the figure, and a muscle preparation interpolated into each circuit. Allow the hammer head to fall, and the point *m* to dip into the mercury in X, and the muscle in the circuit II, passing in the direction *c*, *d*, *h*, *i*, *m*, *x*, *y*, will receive the *closing* shock, while that in the circuit, marked I, *t*, P, *r*, *u*, will receive the *opening* one, because the circuit is broken by the forcible separation of the lever P from the screw *r*. Thus the action of the two shocks may be compared. At Z, underneath the handle of the hammer there is a spring catch which receives it when the hammer falls, holds it securely, and thus prevents vibration.

M'Kendrick's apparatus for measuring tetanus.—This enables us to calculate with readiness the number of distinct galvanic shocks necessary to produce tetanus. It consists of a series of wheels so arranged that if one of them make a revolution in a minute, the next will perform twelve revolutions in the same space of time, the third, 144 revolutions, and so on. Each of these wheels carries on a prolonged axle one or more small wheels of brass, having portions of the circumference cut out at equal distances, and small pieces of ivory interpolated. A steel spring is caused to press against the circumference of one of the wheels while it revolves, and an electric current passing through the instrument is opened each time the spring passes from the brass to the ivory, and closed when it passes from the ivory to the brass. The time occupied by the revolution of the first wheel is calculated by causing it to operate on a spring in connection with a small bell, so that the bell rings at the end of a revolution. Knowing the time occupied by the first wheel, we also know the second revolves twelve times as fast, and the third 144 times, and so on. By means of this arrange-

ment, electric shocks can be transmitted to a muscle or nerve, varying in rapidity from 12 to 3000 a minute. The interval between the shocks may also be calculated to the 1000th of a minute.

Experiments on muscular fatigue.—The only satisfactory way of studying the gradual fatigue of muscle, when stimulated to do a certain amount of work, is to obtain a tracing on a cylinder, or plate of glass, by means of an instrument called a *myographion*, or muscle-writer (*μυων*, a muscle; *γραφω*, I write). Various myographions have been constructed for this purpose, but the most convenient is that of Pflüger, shewn on Plate XX. fig. 5. It consists of a solid wooden stand, S, into which is fixed a strong brass upright, F, carrying upon it a moveable forceps, Z. Into the forceps the end of a frog's femur is securely fixed, the gastrocnemius muscle being attached to it, and hanging downwards. Into the *tendo Achillis* a small hook is inserted, which supports by its other end the lever apparatus *d b*. This lever is a double framework, which moves freely on an axle at the top of the support *a*. At one end of the lever there is an apparatus bearing a stylette, which is employed for making tracings on a plate of smoked glass, P, moving in a frame, B, or still better, a tracing may be obtained on a revolving cylinder. Underneath the lever there is a balance, g, attached by a small hook, *f*, to a swivel apparatus, *c*. Into this balance a weight is placed sufficient to overcome the contraction of the muscle, and the consequent elevation of the lever, when the stimulation is removed. In order to keep the muscle and nerve alive for a considerable time, a square glass case may be placed over it, as shewn in the figure, containing moist blotting paper. By means of this apparatus many interesting experiments may be made, such as the tracing of a muscle stimulated by the opening and closing of a continuous current, the effect of the opening and closing of an induced current, and the effect of muscular fatigue from long-continued stimulation applied directly to the muscle or to the nerve supplying it.

Experiments on Nerve.

The nerve current is demonstrated in exactly the same manner as the muscular current (p. 535). To increase its amount, the nerve should be doubled, and both transverse sections placed in apposition to the cushion (pp. 97 and 169).

Effects of electricity on a nerve.—It is easily demonstrated that

if a current of electricity pass through a motor nerve, it irritates it, and causes contraction of the muscle or muscles supplied by it. It must not be supposed that the electricity is conveyed along the nerve to the muscle ; it only stimulates the former, and calls into action the nerve force which causes the latter to contract. It has been found, however, that the presence and amount of the contraction depends partly on the strength and partly on the direction of the current. This has already been explained theoretically (pp. 169, 176).

Experiments on Pflüger's law of contraction.

To graduate the strength of the stimulation-current, we require ten or twelve small Grove's cells, the zinc surface of each being about $2\frac{1}{2}$ square inches ; and also a special instrument termed a *rheocord*.

The rheocord.—A view of this instrument is seen in Plate XIX. fig. 11. It is for the purpose of enabling us to send a current of definite strength through a nerve or muscle, and to vary this strength at pleasure. When a rheocord is introduced into a galvanic circuit, the current divides itself into two, the one of which we can transmit to the nerve, while the other returns directly to the battery. We have thus a nerve circuit and a battery circuit, and if we interpose resistance in the latter circuit, more electricity will pass through the former. The rheocord consists of a long wooden box. Along one side of this box there are two thick platinum wires, S_1 W_1, and S W, connected at S and S_1 by screws with the two brass plates S_1 with 1 and S with P. At σ σ they pass over a piece of ivory to the screws W and W_1. At one end, on the top of the box, there are six brass plates, 1, 2, 3, 4, 5, and 6, each of which is separated from the other by an interval, which may be readily filled up by a thick ivory-headed brass stopper. Along the surface of the box, and under the platinum wires, there is a piece of brass, Z, capable of sliding backwards or forwards, and having on its surface two hollow cylinders, A, made of polished steel and filled with mercury. These cylinders are pierced by the platinum wires, and the ends directed towards W_1 and W are tightly corked. This brass slide, therefore, forms a bridge between the two platinum wires, which are nowhere else connected by a conducting substance. The current cannot pass from S to S_1 except by going to the nearer cylinder in the direction of the

arrow ——>, and from the other cylinder to S_1, as indicated by the arrow <——. It is, therefore, evident that the amount of resistance offered by the platinum wires to the passage of a current of electricity may be modified by pushing backwards or forwards the brass slide. In proportion as it is pushed towards W_1, W, the resistance evidently becomes greater. Along the side of the platinum wires there is a graduated scale divided into millimetres, which may be used in comparative experiments. In order still farther to modify the resistance at pleasure, there are on the under surface of the cover of the box, in connection with the brass plates 1, 2, 3, 4, 5, 6, a series of wires of German silver, which are represented in fig. 11 by dotted lines, and which, after going up and down in the box, pass from the one brass plate to the other. When all the stoppers are placed between the brass plates, the German silver wires are not in the battery circuit. If, however, we remove the stopper between 5 and 6, the wire connecting 5 and 6 (bracketted at the other end of the figure, and marked X) is brought into the battery circuit, and thus the amount of resistance is increased. The same holds good with regard to the other stoppers. In connection with the brass plates S and 6 there is a short brass upright, each bearing two binding screws, P and Q. The wires from the battery *a a* are connected with the lower binding screws, while those going to the nerve *b b* are attached to the upper.

Mode of demonstrating Pflüger's experiment.—The limb of a frog, having been prepared by dissecting out the sciatic nerve, without injury, is fixed in a pair of brass forceps. The nerve is then stretched over two copper or zinc wires, carefully insulated, and provided with connectors, by which they are attached to two of the cups in a Pohl's commutator, the cross bars being present for the purpose previously described, that of enabling us to transmit a current upwards or downwards in a nerve at pleasure. Two wires are led from the commutator to the upper screws on the rheocord. The lower screws of the rheocord receive wires from the battery, and a key is introduced into the circuit, by means of which we can open or close it at pleasure. The connections having been thus made, we endeavour in the first place to stimulate the nerve by as weak a current as possible. This is effected by using one small Grove's cell and having all the brass stoppers of the rheocord in their places. By this arrangement there is almost no resistance in the battery

circuit, and consequently a weak current is transmitted to the nerve. By means of the commutator, also, we are enabled to transmit the current along the nerve either in an upward or downward direction, and the result is to be observed on opening and closing the key. The strength of the current is now to be increased by using two or perhaps three of Grove's cells, and by removing one or two of the brass pegs in the rheocord. On opening and closing the key, and by moving the commutator, we now observe the effect of a medium current transmitted upwards or downwards along the nerve. The effect of a strong current is shewn by using five, six, or eight of Grove's cells, and by removing all the pegs in the rheocord. By carefully graduating the strength of the current, and operating upon at least $1\frac{1}{4}$ inch of healthy nerve, the results described at page 172 may usually be obtained; but occasionally, from the fact that the nerve is irritable in frogs which have been long kept in confinement, it is difficult to obtain contractions in the order described by Pflüger and others.

Pflüger's experiment to shew that the nerve force accumulates intensity as it advances.—This remarkable fact may be demonstrated by stimulating a nerve close to the muscle, or at a short distance from it, when it will be found that a current too weak to cause contraction of the muscles when sent to a point close to the muscle, will cause powerful contraction when transmitted to a point at a distance from the muscle (p. 175). To succeed with this experiment, it is necessary to use a large frog so as to obtain a long nerve. This nerve is stretched across two pairs of wires, the wires in each pair being placed close together, and each pair being also separated by a distance of about an inch. The wires are placed in connection with a Pohl's commutator, to which also are attached wires from the secondary coil of an induction machine, a key being interposed in the battery circuit. We are thus enabled to transmit the current either near to, or at a distance from, the muscle; and by diminishing or increasing the distance between the primary and secondary coil of the induction machine, we can graduate the strength of the current. The best method of making the experiment is, in the first instance, to remove the secondary from the primary coil to such a distance that no effect is produced when the nerve is stimulated either close to, or at a distance from, the muscle. Having then placed the commutator so that the current

will be transmitted along the wires to the portion near to the muscle, the key is opened, and the secondary coil is gradually approximated to the primary, until a very feeble, almost imperceptible, contraction is produced. On now moving the commutator so as to transmit the current to the portion of nerve at a distance from the muscle, a very powerful contraction at once takes place, clearly demonstrating that the same amount of stimulus produces a greater effect when applied to a nerve at a distance from, than when applied near to, the muscle.

Experiment to determine the rapidity of the nerve current.

This problem, which has received the attention of many physiologists, has now been satisfactorily solved by the labours of Helmholtz and Du Bois-Reymond. The necessary instrument is termed a *myographion*, of which there are several varieties, but the one generally used is that employed by the two distinguished physiologists just mentioned. A sectional view of this instrument is seen, Plate XX. fig. 1, and the arrangement of the apparatus for the experiment, will be understood by referring to the diagrammatic sketch in Plate XX. fig. 3.

Mode of calculating time by tracings on a revolving cylinder.—Before describing the Myographion, the student must understand this important method, which is applied to many experiments in practical physiology. Suppose a cylinder, worked by clockwork, or even steam power, makes one revolution in a minute, and its surface is divided into sixty equal parts by sixty vertical lines at equal distances from each other, the distance between two lines evidently represents in time one second. By measuring the circumference of any cylinder, and by observing the time occupied by one revolution, we can thus calculate the time represented by the distance between any two points in the circumference. This principle, which we owe to Th. Young, 1807, is taken advantage of in the myographion. It is necessary that all revolving cylinders go smoothly, and at a uniform rate. This is attained by a fly-wheel, or a centrifugal apparatus attached to it.

Description of the Myographion.—The myographion, although a very complicated instrument, consists essentially of three parts: 1st, arrangements for holding a muscle having the sciatic nerve attached, the latter being connected with electrodes; 2d, clockwork for moving a revolving cylinder with a certain measured

rapidity; and 3d, arrangements for stimulating the nerve at the particular moment when the cylinder has reached a known velocity.

(1.) The muscle consists of the gastrocnemius (figs. 1 *j* and 3 *b*) attached to the femur, and having the sciatic nerve in connection with it. Into the *tendo Achillis* is inserted a small hook, from which is suspended a long thin iron wire, *k*, the latter being in connection with a lever apparatus, *l l*. This lever, the fulcrum of which is on the top of the pillar F, is balanced behind by a movable weight, *m*, and has at its other extremity a rectangular arm, O, bearing the stylette P. This stylette is opposite the cylinder B, and it is evident, on examining the plate, that if the muscle contract, it will elevate the lever, and the stylette P will make a mark upon the cylinder. At the top of the figure will be seen various contrivances for accurately adjusting the muscle with reference to the lever. The strong brass pillar E supports a square glass chamber, in which the muscle is placed, and in which it can be kept living and moist for several hours, by placing on the inner surface of the glass a few bits of blotting paper dipped in warm water. The floor of this chamber is made of vulcanite, and is perforated by a hole between *r* and *r*, so as to permit the passage downwards of the iron wire *k*, connecting the *tendo Achillis*, with the lever. When the apparatus is being used, in order as much as possible to exclude air, and keep the muscle moist, the hole in the vulcanite floor is almost closed by two semicircular pieces of glass, *r r*, having a small excavation on the straight border of each, so that, when in apposition, only a small round opening is left. The brass forceps may be elevated or depressed by moving them upwards or downwards in the socket marked *c*. This socket has a universal movement in the air-tight box *i*, *e*, *g*, and may itself be regulated by the screws *h* and *e*. Into the bottom of the moist chamber we have usually four double binding screws, three of which are seen in Fig. 1, marked S, S, S, and S′, S′, S′. These are for the purpose of attaching wires to the muscle within the moist chamber.

(2.) The motive power of the clock-work is a heavy weight attached to a strong cord, *a*, wound round a drum, and which passes over a pulley. The situation of the clock-work is seen at A, and on its surface there is a small dial, *b*, so regulated, that when the machinery is in operation, the hand moves one degree for every ten revolutions of the cylinder.

(3.) The most important part of the apparatus is the arrangement for stimulating the nerve exactly at the moment when the cylinder has reached a certain definite velocity. That velocity is usually fifteen revolutions in the second. This part of the apparatus will be best understood by referring to Plate XX. fig. 3, while at the same time further attention is given to Fig. 1. Underneath the lever, and securely fixed to the stand D D of the instrument, there are two brass uprights, Fig. 1, *t*, and Fig. 3, *k*, each bearing a rectangular arm, and each having a binding screw. The two arms come into close apposition with each other at Fig. 3, *g*, but do not touch, the connection between them being established by a strong steel spring attached to one of them. This bridge-like part of the apparatus is introduced into the battery circuit, which also includes the primary coil of an induction apparatus. This will be understood by referring to Fig. 3, where we see *l* the battery, *m* the primary coil of the induction machine, *k* and *i* the piers of the bridge, and *g* the steel spring establishing a connection between them. It is evident, therefore, that when the steel spring completes the bridge, the galvanic current from *l* will pass to *m*, thence to *k*, thence along the bridge *g*, and from thence by the wire *i* back to the battery *l*. But if the bridge *g* be broken by the elevation of the spring, it is also evident that, in accordance with the principles of induced or Faradic electricity, at that moment a secondary current will be induced in *n*, the secondary coil, which secondary current is employed in stimulating the nerve.

The next part of the mechanism is that by which this is accomplished exactly when the cylinder attains a velocity of fifteen revolutions in the second. It is done by means of a rectangular arm of brass marked in Fig. 1 $\pi\pi$, and in Fig. 3 by *h*, bearing at the end farthest from the cylinder, and immediately underneath the spring, a quadrant or arc of brass. When this arm is pushed to the left side, it will be found that the quadrant moves through a distance of a quarter of a circle and elevates the steel spring. It is, therefore, necessary that the arm be pushed over at the proper moment, so as to break the primary circuit. For this purpose, there is an ingenious application of the principle of centrifugal force. Underneath the cylinder B, but on the same steel axis, L L, there is a round brass box, C.

This box, therefore, revolves with the same rapidity as the cylinder, and any two points on the surface of the cylinder and box will always be in the same vertical line. On removing it, and looking into its interior, we see two brass weights, *e* and *c*, as represented in Plate XX. fig. 2, one of which, *e*, is immovably fixed in its position, while the other, *c*, is loose, but in connection with a steel spring, *d*, which keeps it in its place. Connected with the weight *c* there is a curved steel spring or catch, *b*, having at the end of it a hook, which is fixed into a notch on a piece of steel, *a*, bevelled at the point. This piece of steel, *a*, we may term the "out-springer," because, if not restrained by the steel spring *b*, it would tend to dart outwards in the direction of the arrow ⟵, the motive power being the wire spring coiled round the end next *d*. When, however, the box is rotated by the machinery, the weight *c* being movable, tends, by centrifugal force, to pass outwards towards the circumference of the box, in the direction of the dotted lines ⁘, and when it reaches the side of the box, which only takes place when the latter is making fifteen revolutions in the second, the spring *b* has been so altered in position that its catch is removed from the out-springer *a*. This latter being thus released, darts out, and strikes against the end of the rectangular arm already described, pushes it to one side, and thus breaks the bridge by elevating the steel spring *g*, Fig. 3. When this occurs, as already explained, there is an induced current in the secondary coil *n*, Fig. 3, which is transmitted to the nerve.

There is still another part of the instrument to be described, that by means of which the stylette is applied to the cylinder at the proper time. It is evident that it must not be allowed always to be in contact with the cylinder, because it would, in the first place, by friction, interfere with its velocity; and, in the second, a number of indistinct marks would be produced on it, which would injure the proper tracing desired. This is avoided by keeping the stylette within a short distance of, but not touching, the cylinder until the moment of time (the fraction of a second), *immediately after*, the out-springer has come out of the brass box. The arrangement for doing this consists of a bar of brass having the shape ⌐——⌐ (Plate XX. fig. 1, *v'* and *v,*), placed above the brass box, and to the right of the common axle, L L, of the cylinder and the box, having the one end bent at right angles upwards, and the other

downwards, v' and $v_,$. This piece of brass v' $v_,$ moves on an axle τ τ''. The right hand extremity of $v_,$ $v_,$ supports a lever above it, a moving on a fulcrum, τ ρ to the end of which, p, there will be seen a very fine thread passing downwards to φ the end of a balance, V. To the other end of this balance, a short rod terminating at ε (near the base of the pillar F), has attached to it another thread which ascends to v, and there passing over a small pulley, goes onwards to be inserted into the end of the stylette P. When, therefore, the lever is supported by the piece of brass v' $v_,$ the thread is kept tense, and thus the stylette is prevented from touching the cylinder, and the rectangular arm is on a higher level than the end of the out-springer. (See Fig. 1, ρ and ϖ.)

But if we now examine the figure, we shall see near the axle bearing the box and cylinder, a small triangular projection, β. This is the other end of the out-springer (Fig. 1), and α (Fig. 2), projecting through an opening in the lid of the centrifugal box. When, therefore, the out-springer leaps out, the end next the axle β first comes against the piece of brass v' and $v_,$ knocks it from under the lever a, so that the thread is relaxed, and the stylette comes into contact with the cylinder. Not only so, but the same relaxation of the thread allows the rectangular arm to fall down vertically to a level with ρ the end of the out-springer. This occurs immediately after the out-springer has passed the end of the arm, so that when another revolution is made, it strikes against the end of the latter, and thus breaks the bridge in the primary circuit, by elevating the steel spring at g in the manner already described (Fig. 3).

Mode of making the experiment.—Having now explained the mechanism of the instrument, we must next describe the arrangements for the experiment, with the aid of the diagram, Fig. 3. Here we have a muscle, b, attached to a femur, which is held securely by the forceps a r. In connection with the muscle, we have the nerve c stretched on two pairs of wires at a certain distance from each other, say two inches. The problem is to find the length of time the nerve-current occupies in passing from q to r. The *tendo Achillis* is attached by a hook to the lever e, bearing a stylette pressing a cylinder, which is carefully smoked in the flame of a turpentine lamp. On the same axle, and underneath the cylinder, is the centrifugal box f. The primary circuit is now completed

as follows: A wire is carried from the positive pole *k* of the battery, *l* to *m*, the primary coil of an induction machine. From thence it passes to *k*, one of the piers of the bridge, thence along the bridge, traversing the steel spring *g*, and from thence along the wire *i*, back again to the battery *i*. The secondary coil of the induction machine is now connected with Pohl's commutator at *n''*, *n''*, from which two wires near *o*, 1, 2, pass to the portion of nerve at *q*, at a distance from the muscle, while those near *p*, 3, 4, pass to *r*, close to the muscle. By moving the commutator we can thus transmit the induced current from the secondary coil either to the portion of nerve at *q* or at *r*. The arrangements having thus been made, the commutator is placed so as to send the current first to a portion of nerve at a distance from the muscle, as shewn in Fig. 3 at *q*. The clock-work is now set in motion; and when the cylinder has acquired a velocity of fifteen revolutions in the second, a sharp click is heard, caused by the out-springer striking away the rectangular arm, and the muscle at that moment contracts. We now reverse the commutator, so as to stimulate the nerve near the muscle (Fig. 3, *r*), and we arrange the myographion for another tracing. This is done by gently pushing in the out-springer by a little button seen on the side of the brass box, elevating the lever *a*, and introducing beneath it the piece of brass *v'*, *v*,, so as again to remove the stylette from the cylinder. We also readjust the rectangular arm, by elevating it. It will be found that the arm, when struck away, is firmly held down by the spring catch *β*, so as to secure the muscle receiving only *one* induction shock. Everything being again ready, the operation is repeated in the same way, and a tracing obtained of the muscular contraction caused by stimulating the nerve near the muscle. The smoked cylinder is now carefully removed, when there will be found on it a slight tracing like what is shewn in Plate XX. fig. 7, but in much more delicate lines than there represented. Here we have first a basement line, *a b*, and two curved lines proceeding upwards, the one, *c e*, being produced by the contraction of the muscle when the nerve was stimulated close to it, while the other, *d f*, represents the contraction when the stimulation was applied at a distance from the muscle. The distance, then, between the points, *c d*, at which these curved lines leave the horizontal line, indicates the length of time the nerve-current took in passing from the point stimulated at a distance

to the point stimulated near the muscle, that is, along a length of two inches. It follows that as we know the circumference of the cylinder and its velocity, there is no difficulty in calculating the time it represents.

Calculation.—The data given are—(1.) Length of nerve examined, two inches; (2.) Distance between the commencements of the two curved lines, 1-6th of an inch; (3.) Circumference of cylinder, six inches; and (4.) velocity of cylinder, fifteen revolutions in a second. $15 \times 6 = 90$, the number of inches of cylinder which pass before the stylette in one second. 90÷1-6th=1-540th of a second, that is, the distance 1-6th of an inch on the cylinder represents in time the 1-540th of a second. Now the 1-6th of an inch on the cylinder indicates the length of time the nerve-current took in travelling along two inches of nerve. Therefore the nerve-current travels two inches in the 1-540th of a second. But two inches are the 1-6th of a foot; therefore the current will travel a foot in 6-540th of a second, that is, 1-90th of a second, or 90 feet in one second.

There are many other details in the management of this instrument which can only be understood by careful study and practical manipulation.

Other myographions have been used by Du Bois-Reymond, Æby, and Marey, but the one now described is the most accurate. No one can study it without being profoundly impressed with the great amount of ingenuity and skill displayed in its construction.

Experiments on the Circulation.

Before making any experiments on the circulation, the student should study the phenomena of hydrostatics and hydrodynamics described at p. 114, and the phenomena produced by injecting water through non-elastic and elastic tubes.

1. *Non-elastic tubes.*—For this purpose a glass tube, having a diameter of 1-8th of an inch, and about eight feet long, may be employed. On driving water through this tube by means of an india-rubber syringe, it will be found that the fluid will pass from the other end of the tube in a series of jets, or *per saltum*. In this case, the tube being almost non-elastic, there is no wave-like motion. If, however, an india-rubber bag be attached to the end of the glass tube farthest from the syringe, it will then

be seen that the jet-like efflux of the fluid has now been converted into a more or less uniform flow.

2. *Elastic tubes.*—In order to demonstrate the effect produced upon a current of fluid forced through an elastic tube, about eight or ten feet of good india-rubber elastic tubing is required. On injecting water through it, the fluid will not pass out in a series of jets, or *per saltum*, but with a uniform flow. It should now be demonstrated that the elasticity of the tube has the property of converting the impulse of the syringe into a wave-like motion, which may be done by means of an apparatus devised by Marey, termed a *triple sphygmograph.*

Marey's triple sphygmograph.—This consists of an arrangement of three light wooden levers of equal length moving on a delicate fulcrum placed near one end of each lever. These levers are carefully fixed, by means of screws, to a wooden upright, so that each lever is about $1\frac{1}{4}$ inch from the other, but in the same vertical plane. To the free end of each lever there is attached a small pen or brush, so that when brought into contact with a revolving cylinder, any movement they make is registered in the form of a curve. The apparatus being thus arranged, the india-rubber tubing is brought under the lowermost lever, about one foot from the syringe, and is placed in a grooved brass support, so that the lever rests lightly upon it. About four feet of the tube is now carried round the rings of two retort stands, fixed on their supports about a foot in height, and then it is brought under the second lever in the same manner. Other three feet of tube are carried over the rings of the retort stands, and after passing under the third lever, the end of the tube is conveyed into a vessel for receiving the water. Thus we have three levers placed at different distances on the india-rubber tubing. On now carefully bringing the free ends of the levers against a smoked cylinder, caused to revolve by clock-work, and forcing water through the tube at regular intervals from a syringe, we obtain on the cylinder three exactly parallel and comparable traces. The tracing produced by the lever next the syringe will shew a series of waves with a bold vertical line, a somewhat abrupt summit, and a gradual descent. That obtained by the second lever will shew waves with a more gradual ascent, a more rounded summit, and a gradual descent; while the tracing made by the third lever will manifest only minute waves. Thus

it may be demonstrated that the original force of the impulse, and the extent of the vertical movement of the lever diminishes with the increase of distance from the impelling organ, whilst the time of impulse remains the same. Other experiments to shew how the pulsatile is converted into a uniform flow of fluid, as in the capillaries and in aneurisms, with rigid or elastic coats, may also be demonstrated with this instrument.

Experiments on the pulse.

These may be made by means of various instruments.

Vierordt's sphygmograph (from σφυγμὸς, the pulse; and γράφω, I write).—The first form of this instrument was made by Vierordt, and consisted of a lever capable of being accurately balanced by shot or sand placed in a cup or cups situated near one end. From the lower surface of the lever, a little rod descended furnished with a small button, which was allowed to rest upon the artery. This rod was attached to the lever, close to the fulcrum, so that a slight movement of it produced an extensive movement of the long arm of the lever. This latter was brought in contact with a revolving cylinder. Tracings were thus obtained, but the inertia of the lever was too great for the force of the circulation, and the periods of contraction and dilatation of the ventricle were not accurately marked.

Marey's sphygmograph.—The principle of this instrument is exactly the same as that of Vierordt's, but it has the advantage of having a very light lever, and of being altogether a much less cumbrous instrument. A view of this instrument is seen in Plate XXI. fig. 13, in which it is represented as attached by bands to the left arm, so that a tracing may be obtained of the pulsations of the left radial artery. A diagrammatic view of the essential parts of the instrument is seen in Fig. 14. It consists of a very light wooden lever, Fig. 14, *a b*, moving on a fulcrum seen at *k*. Underneath this lever there is a fixed bar, *c d*, to the under surface of which, near *c*, there is attached a steel spring, *e f*, bearing at *f* a button padded with leather or ivory, which rests upon the pulse. From the upper surface of the button *f*, there passes a vertical screw, *f i*, which ascending through a slit in the fixed bar *c d*, bears on it a small steel projection, which when the screw *f i* is elevated, also raises the lever *a b*, in the direction indicated by the dotted lines *g h*. The

lever is brought down to its original position, *a b*, by a delicate steel spring marked *k*. It is evident, on studying Fig. 14, that very delicate movements of the button *f* will produce considerable extent of movement of the lever *a b*, at the point *a*. This point bears at *a* a small pen, slightly bent, which touches the side of a card or plate of smoked glass, or smoked gelatin or mica, which is made to move gradually forward in the direction of the arrows ———> ———> by clock-work (see Fig. 13). The degree of pressure by the button *f* (Fig. 14), on the pulse may be regulated by the screw *i f* (Fig. 14), seen also at *a* (Fig. 13.) The objection to this form of sphygmograph is, that there are no means of accurately measuring the pressure of the button on the pulse. This has now been accomplished, in recent instruments, by means of a screw which regulates the pressure of the spring (Fig. 14) *k*. The head of this screw is divided into equal parts, each division representing so much pressure in grammes-weight, so that in taking tracings of the same pulse at different times, we are enabled to use exactly the same amount of compression upon it. Other sphygmographs have recently been constructed by Mayer & Meltzer, of London, which may be easily adjusted to the wrist by a bracelet, and in which the tracing is obtained on a piece of mica surrounding a small revolving cylinder. For a description of the pulse, and of the meaning to be attached to a sphygmographic tracing, see pp. 216, 217.

The cardiograph of Marey.—A view of this apparatus is seen in Plate XXI. fig. 16, by which Marey was enabled to obtain three tracings simultaneously from the right auricle, the right ventricle, and the pulsation of the heart through the walls of the chest. It consists essentially of (1.) a registering apparatus ; (2.) small oval sacs or ampullæ, made of india-rubber, for receiving the impulse in the vessels or cavities of the heart ; and (3.) an apparatus termed a *tambour* or drum, for communicating the impulse to a lever. The registering apparatus is a cylinder, A, E, moved by clock-work, on which there is enrolled a long band of paper. The levers *le*, *lv*, *lc*, placed one above the other in the same plane, touch the paper by the point of the pen, which terminates each (Fig. 15, *l*, P). The ampullæ for receiving the impulses are seen in Fig. 16, *c*, *v*. These communicate with the tambours by long elastic tubes, containing air, marked *tc*, *tc*, *tv*, which are supported by two iron stands, seen one on each side of A 3. A separate view of

one of the tambours is seen in Fig. 15. It consists of a shallow drum, T, on the surface of which there is placed a thin circular plate of aluminium, *a*. This supports, and is attached to, a short upright in connection with the lever *l*, *l*, P. There is an arrangement seen above E for moving the drum backwards or forwards, and there is also a screw by which we can move the position of the aluminium plate. A short brass tube communicates with the bottom of the drum, or opens into its side, as seen at *b*, to which the elastic tube is fixed, so that the slightest pressure of the air in the drum communicated through the elastic tube produces a movement of the lever. The advantage of this apparatus is that it is easy of application, and its disadvantage is that, in consequence of the great amount of elasticity of the india-rubber drum head, the lever is apt to produce a number of secondary curves instead of one firm, well-defined line. It may be applied to the registration of many other kinds of movement.

The Sphygmoscope of Scot Alison.—This instrument, as its name indicates, is an apparatus for shewing the movements of the pulse to the eye. It is a truncated cone made of brass, the base consisting of a piece of highly elastic india-rubber. To the truncated end of the cone there is a piece of india-rubber tubing passing to a glass tube, bent near the same end to an angle of about forty-five degrees. The apparatus being nearly filled with a coloured fluid, such as infusion of litmus, it is evident that the slightest impulse communicated to the elastic base of the cone, will be at once seen by an elevation and depression of the coloured fluid in the tube.

The Sphygmoscope of Czermak.—This instrument consists of a small rectangular mirror, so fixed by its upper extremity as to move freely when the lower extremity rests upon the pulse. If the instrument be now fixed to the arm so that the free end of the mirror rests on the radial artery, and a strong ray of light be reflected from the mirror upon a vertical scale, the slightest movement of the mirror will be manifested by a great extent of movement of the spot of light on the scale. Thus the movements of the pulse may be exhibited to a large audience.

The Sphygmosphone of Upham.—This apparatus, seen in Plate XXI. fig. 17, is for the purpose of enabling the ear, by means of electric bells, to determine the frequency and rhythm of the cardiac and radial, or femoral pulsations. It may be

divided into two parts: (1.) An arrangement for receiving the impulse, and by means of this impulse, breaking an electric circuit; and (2.) an apparatus for ringing one or other of a pair of electric bells when the circuit is broken.

The arrangement for receiving the impulse from the apex or base of the heart through the thoracic wall, and from any large artery, consists of two small bell-shaped glasses, *l*, *m*, having the mouth of the bell covered over with thin india-rubber. From the other end of the bell we have two elastic tubes which are attached at their other extremity to similar bell-shaped glasses, also closed by india-rubber, marked *i* and *i'*. When these tubes and bells are carefully filled with water, it is evident the slightest impulse received on the india-rubber surfaces of *l m* will be communicated to those of *i* and *i'*. Immediately over *i* and *i'* there are two brass bars moving upon hinges, and having their free ends directed inwards, and resting upon a square piece of brass, as seen at *k* and *k'*. Attached to the under surface of each of these bars there is a round metallic plate, which rests on the india-rubber surface of the glasses *i* and *i'*. By this arrangement, a very slight impulse communicated through the fluid in *l i* and *m, i'*, will elevate the brass bars and break the contact at *k* and *k'*.

The other portion of the apparatus consists of two electric bells, *a* and *b*, which are rung by the hammers *c* and *d*, attached to the keepers *e* and *f* of the two electro-magnets *g*. The current of electricity from two of Smee's elements is conveyed into the apparatus by the wire *p*, passes through both electro-magnets, which, by attracting their keepers *e* and *f*, withdraw the hammer *c* and *d* to a short distance from the bells *a* and *b*. The point of contact between the two electro-magnets, as already explained, is at *k* and *k'*. If, therefore, an impulse be communicated through the apparatus *l i* sufficient to break the contact at *k*, by elevating the brass bar, the keeper of the electro-magnet *g* is released, and the bell *a* will ring, only one stroke of the hammer *c* being given. In the same manner the bell *b* is rung through the influence of *m i'*. By means of this apparatus the interval of time between the cardiac and radial pulsations may be rendered evident to an audience by the different tones of the bells; and as the contact-breaking part of the apparatus may be in Edinburgh, and the electro-magnetic apparatus in London, a demonstration might be given by telegraphic wires to a London

audience of the cardiac and radial pulsations of an individual in Edinburgh.

Experiments to measure the rapidity of the circulation.

This may be estimated roughly by means of two instruments.

1. *Hæmatachometer of Vierordt* (αἷμα, blood; τάχος, speed; μέτρον, measure).—The essential part of this apparatus is seen in Plate XXI. fig. 20. It consists of a rectangular chamber, A B, the sides of which are made of glass. This chamber is furnished with two nozzles, *a* and *b*, for insertion into an artery which has been cut across. In the anterior part of the chamber there is a very light vertically-suspended pendulum seen at *c*, placed close to the point of entrance of the current of blood into the chamber. This will of course move the pendulum from the perpendicular as seen at *e*, and the amount of this deviation will indicate the velocity of the current. To this apparatus is suspended a long lever placed above the box A B, which is moved by a rack and pinion arrangement by the hand of the experimenter, synchronous with the movements of the pendulum. The free end of the lever is provided with a pen or brush, so that a tracing may be obtained on a revolving cylinder. The objection to this instrument is, that the conditions of a square box are different from those of a blood vessel; the inertia of the pendulum has to be overcome, the accuracy of the tracing depends on the quickness of the eye and steadiness of hand of the experimenter, and the whole apparatus is large and difficult of application.

2. *Hæmadromometer of Volkmann* (αἷμα, δρόμος, a race-course, μέτρον).—This instrument is seen in Plate XXI. figs. 18 and 19. It is composed of a U-shaped tube, *d*, *e*, of a given length, having attached to it a scale, graduated in millimetres. The ends of this glass tube fit at *d* and *e* into a brass apparatus, *a*, *b*, having nozzles at *a* and *b*, which are inserted into the cut ends of the artery. This part of the apparatus is furnished with a screw, or stop-cock, bearing a rectangular arrangement of brass tubing, so that by turning it the blood may be caused to flow directly from *a* to *b*, as seen at Fig. 18, or along the U-shaped tube, as seen in Fig. 19. The experiment is thus performed:—The artery is laid bare, and the circulation through two inches of its extent is controlled by two strong spring forceps fixed on it at that dis-

tance from each other. It is then divided, and the nozzles *a* and *b* inserted into the cut ends, and firmly secured by ligatures. The stop-cocks are arranged, as seen in Fig. 18, and the spring forceps being removed, the blood will, of course, flow directly from *a* to *b*. The experimenter being provided with an accurate chronometer, on a given signal the stop-cock is turned, so that the blood will flow along the U-shaped tube, as seen in Fig. 19, and the time it occupies in passing from *d* to *e* is noted. The length of the U-tube being known, the rapidity of the circulation is, of course, thus determined. The U-tube, however, being almost non-elastic, and in no way fulfilling the conditions of a living artery, the determination of the rapidity must be held as only approximative.

Experiments to measure blood-pressure.

1. *Hæmadynamometer of Poisseuille* (αἱμα, blood; δυναμις, power; μετρον, a measure). — This instrument consists of a long U-shaped glass tube, Plate XXI. fig. 21, *a*, *b*, of uniform calibre, and having the inner surface exceedingly smooth. Into the tube some clean mercury is placed, which comes to a level in the two limbs as seen at *d e*. Attached to both limbs there is a graduated scale for registering the oscillations of the mercurial columns. From the limb of the tube at *a*, there passes the curved leaden tube *q l m*, having an air hole at *n*, a joint at *l*, a stop-cock at *o*, and a nozzle placed transversely at *q*. This bent tube fits the U-shaped tube at *a* very accurately, and there is a collar, *m*, which is screwed over the connection so as to make the junction perfectly water-tight. The transverse nozzle at *q*, is represented of larger size in Fig. 21, *b*. It consists of the tube *a b*, Fig. 21, for insertion into the artery, and of the short tube *c* placed at right angles to *a b*, which fits accurately into the end of the leaden tube at *q*. The whole of the leaden tube *q l m*, and the upper limb of the glass tube *a d*, to the surface of the mercury at *d*, is carefully filled with a solution of carbonate of soda, which is used because it has the property of preventing the coagulation of the blood in the nozzle *q*. In accordance with the law that fluids press equally in every direction, the pressure of the blood passing through the nozzle *q*, is communicated latterly through the column of the solution of carbonate of soda to the surface of the mercurial column at *d*. The blood does not flow into the leaden pipe

q l m, but passes straight through the nozzle *q*, and the increase and diminution of pressure is indicated by the movements of the mercurial columns at *d* and *e*. The amount of this pressure is measured by the scales *c*, Fig. 21. If the mercurial column be depressed one-fourth of an inch at *d*, it will be elevated the quarter of an inch at *e;* the total pressure, therefore, being one-half inch of mercury.

2. *Kymographion of Ludwig* (κυμα, wave, γραφω).—This instrument, as seen in Plate XXI. fig. 21, is the apparatus just described, together with an arrangement for registering the movements of the mercurial column. This is effected by means of a very light glass float, *f*, having attached to it a long fine steel wire, *f g*, at the top of which there is a steel rod bearing a stylette, *i*. From the upper end of the rod *g*, a delicate thread passes upwards to the top of the frame *h h*, moves round a small pulley, and suspends a weight, *k*, which acts as an equipoise to the glass float *f*. The rod *g*, bearing the stylette, moves in the same vertical plane, being kept in position by two fine steel wires in the frame *h h*. This apparatus enables us to obtain very delicate tracings, the movement of the mercurial column being communicated to the float, and by the latter to the stylette. To the left of Fig. 21, a revolving cylinder, *b*, is shewn, moving upon an axle, *a c*, and having a stylette, *d*, applied to its surface.

When it is desirable to make an experiment, with the view of determining the amount of blood pressure, the leaden tube must, in the first place, be carefully filled with a solution of carbonate of soda. For this purpose we require several syringes, having nozzles of various sizes. The stop-cock at *o* is perforated through the top, so as to permit the escape of air, and it is furnished with a small stopper for preventing the escape of the fluid. By means of this stop-cock we can allow the blood pressure to be communicated to the mercurial column, or we can shut it off at pleasure. The leaden tube is to be filled by inserting the nozzle of a syringe into the hole in the stop-cock *o*, the latter being turned so as to prevent the escape of the fluid through the nozzle *q*. Thus we fill the tube *l, m*, and usually part of the tube *n, m, d*,—the air escaping by the air-hole *n*. We next introduce the point of the syringe at *n*, and endeavour to fill the remainder of the tube. When we have done so, we must rapidly remove the syringe, and introduce a

stopper into the air-hole *n*, taking care to bring the mercury in the two limbs of the U-tube to a level. The nozzle is introduced into the artery in exactly the same way as already described with reference to the Hæmadromometer. To obtain an accurate tracing, three things must be carefully attended to :

1. The blood must be prevented from coagulating in the nozzle introduced into the artery, and occasionally it may be necessary to remove the nozzle, and to clean it out by a fine wire or bristle, after properly securing the artery.

2. The float must move freely in the glass tube. Before using the apparatus, the tube ought to be carefully dried with a piece of cotton attached to a long wire, and the mercury employed must be perfectly free from dust or moisture. Before using the latter, it should be carefully filtered through a minute perforation in a sheet of white writing paper, and be dried by placing it in a porcelain capsule for five or ten minutes before the fire.

3. The stylette must mark the cylinder with as little friction as possible. This is accomplished by attaching to its point a small finely-pointed brush, which is kept wet with ink of sufficient fluidity ; or by means of a small conical glass, fixed to the stylette, having the apex drawn to a very fine point, perforated and bent so as to barely touch the cylinder. A small quantity of ink is placed in the glass, and passes in a very fine stream through the perforation in the bottom.

In addition to the curve produced by the movements of the recording apparatus, it may be serviceable to have two other tracings made at the same time, one being a horizontal line, drawn by a fixed stylette, and the other a rapid but regular tracing of a series of equally sized secondary curves, produced by another stylette in connection with a magneto-electric apparatus working with great regularity. In the event of numerous experiments being required in any particular investigation, the whole apparatus should be permanently fixed to a table, which is also used as the operating table, and on which the animal lies.

Experiments on Respiration.

1. *Expiration of carbonic acid by the lungs.*—This may be readily demonstrated by breathing through a glass tube into lime-water. The lime-water becomes milky from the formation of insoluble carbonate of lime. The amount of carbonic acid expired may also be determined by causing an individual

to breathe for an hour air which has been carefully purified by passing it through caustic potash. He must inspire this pure air and expire through a solution of caustic potash, the strength of which is known, and which may be weighed. The increase in weight, due to the carbonic acid, indicates the amount of the latter; or the amount of carbonic acid may be calculated, by the chemical rules of equivalence, from the amount of carbonate of potash formed.

2. *Expiration of aqueous vapour by the lungs.*—The amount of aqueous vapour may be estimated by breathing air (dried by passing through sulphuric acid) through an apparatus consisting of several U-tubes containing chloride of calcium, or pumice-stone steeped in sulphuric acid. The increase in weight will indicate the amount of aqueous vapour expired in a given length of time.

3. *Mode of measuring the quantity of air in inspiration and expiration.* (See pp. 228, 229.)—This is done by means of instruments termed *spirometers.*

(1.) *Spirometer of Mr Hutchinson* (*spiro*, I breathe; *μετρον*). —This is essentially a gasometer, consisting of an outer cylinder, having introduced at its base a pipe, leading from a mouthpiece, and rising in the centre of the cylinder nearly to its brim. Into this cylinder another one is inverted, which is carefully balanced by two cords passing over two pulleys, and suspending two weights. When air is forced into the tube already mentioned, and the outer cylinder contains a certain quantity of water, the inner cylinder rises. The amount of air is indicated by a scale, graduated into cubic inches, which is attached to the inner cylinder, and consequently rises with it, the amount being marked off by an index fixed to the outer cylinder. The inner cylinder is provided with a stopper, by removing which, and forcing down the cylinder, we are enabled to expel the air. This instrument does not give accurate scientific results—the muscular strength of an individual influencing the amount of air forced from the lung, independently of the pulmonary capacity.

(2.) *The Anapnograph of Bergeon and Kastus* (*ἀναπνεώ*, to draw breath; *γράφω*).—The principle of this instrument is quite different from that of Mr Hutchinson. It is seen in Plate XXI. fig. 22. It consists of two parts: first, an arrangement of clock-work for carrying a sheet of ruled

paper, P C, placed in a brass box, M N ; and second, of another apparatus consisting of a rectangular box, R, into one side of which we have fixed an india-rubber tube, terminating in a cover for the nose, as seen at O. The other side of the box is quite open, as at V. A section of the box is seen at C. In its interior there is a vertical plate of aluminium, moving freely on a hinge, at the bottom of the box. This plate, seen in C, 3—2 acts as a valve during inspiration and expiration through the box. In inspiration the valve is drawn towards the nose, as indicated by the dotted line in C, 3—4 ; and in expiration it is forced in the opposite direction. To the edge or border of this valve there is attached a long light lever, K, having at its free end a pen, P, which, as the lever is moved by the valve, makes a tracing upon the paper. It is important to observe, as will be seen on examining C, that when the valve is forced from the nostrils in expiration, the pen point will move in the opposite direction ; and the reverse holds good in the case of inspiration. The whole of the tracing on the left of the median line of the paper, therefore, represents the curve of expiration, while that on the right represents that of inspiration. Thus we obtain a tracing of the inspiratory and expiratory curves. The amount of air of inspiration and expiration is calculated by having the paper carefully divided into squares, so that in ordinary respiration sixteen squares represent half a litre of air, while in forced respiration, four squares represent only half a litre. Thus by counting the number of squares within the curve, we obtain a knowledge of the amount of air. For ordinary respiration the apparatus is as shewn on Fig. 22, and the pen hangs loosely. In this condition forced respiration would drive the pen point beyond the margins of the paper, and perhaps damage the instrument. To obviate this, by turning the small button B, seen near the top of the box, pushing it downwards in a little slit, and again fixing it, the lever is rendered much less easily moved. This instrument is easy of application, and is more accurate than that of Hutchinson.

Experiments on Sight or Vision.

The practical experiments which may be performed with reference to sight are so numerous that we can only select a few of the more important.

1. *Inversion of the image upon the retina.*—This may be illus-

trated by the student examining the inverted image upon the ground glass plate in an ordinary photographic camera. It may be also done by taking the eye of an ox newly killed, or still better, that of an albino rabbit, carefully separating the sclerotic from its posterior surface, and fixing it in a hole in a shutter, the pupil being directed forwards, while the observer is in the dark room examining the retina.

2. *Action of the muscles of the eye-ball.*—The *ophthalmotrope of Reute* (οφθαλμος, the eye ; τροπη, a turn). By means of this instrument, seen in Plate XXI. fig. 23, we are enabled to study the actions of the muscles of the eye-ball. It consists of a wooden box, *h*, supported by levelling screws, *i k*. From the surface of the box there rises a brass pillar, *g*, bearing a framework in which there are mounted the accurate models of two eye-balls. These latter consist of box-wood frames, having passing through their centre a brass tube, bearing on its anterior part a glass, representing the cornea, behind which there is a diaphragm, like the pupil, while its posterior end consists of a disc of ground glass, which stands for the retina. The six muscles acting upon each eye-ball are represented by as many delicate silk cords, accurately fixed to their proper position on the eye-ball. From thence they pass backwards through two brass plates, *c d*, then over a number of ivory pulleys downwards towards two scales, *f f*. The back of one of these scales is seen in Fig. 23, A. Each cord has attached to it a small piece of tinfoil, which serves as a pointer. The silk cords are ultimately attached to a roller in the box *h*. The peculiar position and direction of the superior oblique muscles of the eye-ball are imitated by a movable arm seen near the inner surface of the eye-ball *a*. By turning with the fingers the eye-balls of this ophthalmotrope, the action of the various muscles may be observed, and the angles formed by the lines of direction of the muscles measured. Numerous other experiments may be performed with it. By placing a wax candle eight or ten feet in front of the ophthalmotrope, the position of the inverted images on the retinæ, corresponding to any given direction of the visual axes, may also be studied.

Measurement of the Curvatures of the Cornea and Lens.

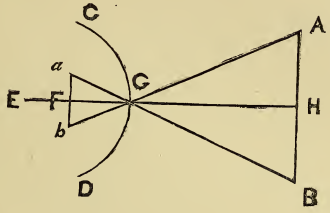
The cornea being a transparent structure, acts both as a lens and as a convex mirror. It acts as a lens by refracting to a

slight extent rays of light passing through it, and it acts as a convex mirror by reflecting rays of light from its surface. It is the latter property that is taken advantage of in the method of making accurate measurements of its radius of convexity.

Formation of images by convex mirrors.—The image produced by a convex mirror is an erect image apparently placed behind the mirror. This will be understood by referring to Plate VIII. fig. 21. Here A P is a convex mirror, and C is the centre of the circle of which C D is the radius, and A P is an arc. If the eye of the observer be placed at E, a reflected and erect image of the arrow M N will be seen at *m n*, but reduced in size. Of the numerous rays of light reflected from the surface of the mirror, only a few can enter the eye, and those which do, such as D E, F E, G E, and H E, are so reflected that the angles of incidence and reflection are equal. The ray M D is reflected in the direction D E, the angle of incidence M D N being equal to the angle of reflection N D E. The same is true of the rays M F, N H. By carrying back the rays E D, E F, they will be found to meet at the point *m*, and they will appear to an eye placed at E as if they had come from their focal point *m*. In the same way the rays E G, E H, will apparently issue from *n*, —all the points composing the image *m n* being foci conjugate to the points composing the object M N. The small image *m n* will therefore be the virtual image of M N. By drawing the lines M C, N C, it will also be found that the virtual image *m n* is always within those lines, hence the image is erect and always smaller than the object. It is important also to recollect that the size of the image *m n* is to the size of the object M N as the distance of the image from the centre of the mirror *m* C is to C M, the distance of the object. The image *m n* will recede from the surface of the mirror, as the object M N recedes from it, and when the object M N is indefinitely distant, as it often is in the case of objects placed before the cornea, the image *m n* will be situated about half-way between the mirror and C, that is at a point corresponding to half the radius of convexity. It follows also from this, that the greater the degree of convexity of the surface of the mirror, the smaller will be the virtual image, a fact which may be easily demonstrated by comparing the sizes of the reflected images in convex mirrors of different degrees of convexity.

Formula for calculating radius of curvature of cornea.—When

we apply these principles to the cornea, we find that it acts as a convex mirror, having a virtual image behind it at a point situated at a distance of half the radius of its convexity. The size of the image must be measured, and from its size the radius of curvature may be calculated thus :



Let C D in the above Figure be the cornea, and E F H its optical axis. The object A B, placed before it will be reflected by its surface, so that its virtual image will be *a b*, placed at F, that is at a distance of half the radius E G. Draw the lines A *b*, B *b*. The object A B will be to the image *a b* as the distance H G is to G F, that is half the radius. Let R = the radius—

$$\text{A B} : a\,b \;::\; \text{H G} : \text{G F (that is half R)}$$

$$\tfrac{1}{2}\,\text{R} = \frac{\text{H G} \times a\,b}{\text{A B}}$$

$$\text{Or R} = 2\left(\frac{\text{H G} \times a\,b}{\text{A B}}\right)$$

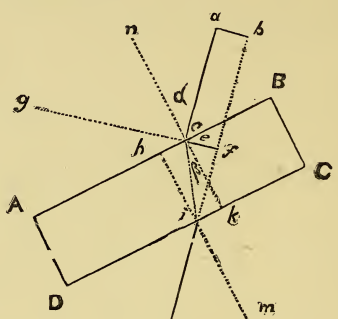
Thus let A B = 1000 mm.; *a b*, 1 mm.; and H G, 3800 mm.; what is the radius of curvature ?

$$\tfrac{1}{2}\,\text{R} = \frac{3800 \times 1}{1000} = 3{\cdot}8$$

$$\text{R} = 3{\cdot}8 \times 2 = 7{\cdot}6 \text{ mm.}$$

Optical principles of the ophthalmometer.—The instrument by means of which we measure the size of the reflected image on the convex surface of the cornea or lens is termed the Ophthalmometer (*ὀφθαλμος*, the eye ; *μετρον*, a measure). This ingenious instrument was invented by Helmholtz. In order to understand its practical application, it is necessary, in the first instance, that we examine the optical principle on which it is constructed. When a ray of light falls perpendicularly on the surface of a glass plate, it passes through it without under-

going any refraction. If, however, the plate be held obliquely to the direction of the ray, as seen in the accompanying figure, we obtain a different result.



Here A B C D in the above figure represent a plate made of flint glass, having the ray *a c* impinging upon its surface. It is refracted in the direction *c i*, and on passing again from the flint glass into the air, it is a second time refracted in the direction *i l*,—*i l* being parallel with *a c*. Draw a line perpendicular to A B, namely *n c*, and continue it to *k*. The angle *a c n* is equal to the angle *m i l*, being produced by parallel lines falling on parallel surfaces. The angle α bears a ratio to the angle β,—the angle of refraction. The index of refraction of flint glass is 1·6. Hence the sine of the angle $\alpha = 1{\cdot}6$ (sin β). Consequently, if the eye be situated at *l*, the point *a* will not be seen at *a*, but at *b*, in the direction of the line *l f b*. The glass plate, therefore, effects a *displacement* of the point *a* to the right, and to an extent indicated by the length of the line *a b*. As yet we do not know the length of *a b*, but it may be represented by the line *c f*, which is equal to *a b* by parallel lines. The line *c f* we will term *e*. In the triangle *c f i*, *c f* is opposite to the angle *c i f*, and *c i* is the hypothenuse, therefore—

$$\frac{e}{c\,i} = \sin\, c\,i\,f.$$

and therefore

$$e = c\,i \cdot \sin\, c\,i\,f.$$

But *e* has not yet been measured, neither do we know the length

of $c\,i$, nor the angle $c\,i\,f$. We must now find $c\,i$. This line is, as the figure shews, the hypothenuse of a triangle $c\,i\,k$; and one of the sides of this triangle, $c\,k$, is equal to the thickness of the glass plate, which we will term P. The line $c\,k$ is adjacent to the angle $c\,i\,k$, and hence we get

$$\frac{\mathrm{P}}{c\,i} = \cos\ \beta,$$

by multiplying $c\,i$, we have

$$\mathrm{P} = \cos\ \beta \cdot c\,i,$$

wherefore

$$c\,i = \frac{\mathrm{P}}{\cos\ \beta.}$$

Now we know that P equals the thickness of the glass plate, and that the sines of the angles α and β are in a known ratio. But by substituting the value of $c\,i$ in the previous equation, we have

$$e = \frac{\mathrm{P}}{\cos\ \beta} \sin\ c\,i\,f.$$

We see now that the $c\,i\,f$ equals $h\,i\,f - h\,i\,c$; but $h\,i\,f$ equals the angle of incidence α, in that their sides are parallel; and $h\,i\,c$ equals the angle of refraction β, because they are alternate angles. Therefore $c\,i\,f$ equals $\alpha - \beta$, and $\sin\ c\,i\,f = \sin\ (\alpha - \beta)$. We have therefore the following formula: (e representing the amount of displacement of the point a towards b and P the thickness of the plate).

$$e = \mathrm{P}\ \frac{\sin\ (\alpha - \beta)}{\cos\ \beta.}$$

But as there are two such plates in the ophthalmometer, we have the complete formula, in which E will equal the total displacement.

$$\mathrm{E} = 2\,\mathrm{P}\ \frac{\sin\ (\alpha - \beta)}{\cos\ \beta.}$$

The use of the ophthalmometer, therefore, is to supply us with the angle α, and as we know the thickness of the glass plates, and the index of refraction between air and flint glass (namely 1·65), by applying the above formula, the amount of lateral displacement may be ascertained. Suppose the thickness of the glass plate to be ·325 mm., the index of refraction 1·65, and the angle of incidence 6°, we find, by the use of logarithmic tables, that $\beta = 3°\ 37'$, very nearly. Therefore $\alpha - \beta = 6° - 3°$

37′ = 2° 23′. This gives, on referring to the tables, e = 0·027 mm. very nearly.

Description of the ophthalmometer.—This instrument consists of a telescope suitable for short distances, part of which is seen in Plate XXI. fig. 24, *c*. In front of the telescope there is a square brass box, *a b b*, having the inner surface blackened. The box has a circular opening near *b b*, which is usually closed with a plate of very thin glass. In the interior of this box there are vertical plates of flint glass *b b*, fitted into frames, and moving the one at an angle with the other, by means of a rack and pinion movement, on a circular disc or wheel seen in Fig. 24, *c*, which is set in motion by turning the screws seen on the top and bottom of the box, the lower one being marked *d*. In Fig. 24, *f*, we see the inner surface of the bottom of the box with the circular wheel just alluded to. In connection with each pinion, and placed on the outside of the box, there are two circles made of steel, and carefully graduated into 360 degrees. These circles are upon the same axis as that on which the glass plates in the interior of the instrument revolve, the upper circle corresponding to the one plate, and the lower to the other plate. There is a fixed vernier at the point *a*, so that by observing the number of degrees on the circle opposite the zero of the vernier, we read off the number corresponding to the obliquity of the plate, or in other words, the number of degrees formed by the angle of incidence, namely, α. In making accurate observations, a number of readings should be taken on both graduated scales, and the differences between these readings should never exceed the one-tenth of a millimetre. If they do, the instrument is not in proper order, owing to the plates not moving at equal rates, or owing to a flaw in the glass.

Mode of using the ophthalmometer.—The first requisite for ophthalmometric observations, is a room having the walls blackened, and from which all sunlight can be excluded. The ophthalmometer is placed at a distance of ten feet from the eye under examination, and on a level with it. The object to be reflected on the cornea, until recently, was the distance between three candle flames placed beside the experimenter, two being on his right hand and one on his left. Helmholtz has, however, now substituted for this an apparatus consisting essentially of three small *rectangular mirrors* fixed by universal joints to a graduated wooden rod about four feet long. The

distance between the mirrors may be regulated by sliding them in moveable sockets along the rod. In the centre of the rod there is a circular movement round a graduated scale, so that the rod may be placed vertically, obliquely, or horizontally, according as it may be desirable to obtain images in a vertical, oblique, or horizontal meridian of the cornea. This mirror-apparatus is screwed to a table immediately in front of the ophthalmometer. A candle flame is now placed on the right or left hand side of the eye to be examined (for the right eye on the right side and for the left eye on the left), as near it as possible, and on the same plane, a dark screen intervening, so as to protect the eye from the glare of light. The experimenter now throws, by means of the mirrors, a reflection of the light from each mirror upon the eye. He then directs his own eye to the telescope of the ophthalmometer, and by carefully focusing the instrument, and directing it to the eye under observation, he sees three minute specks of light, thus * * * on the cornea. The vernier of each scale of the ophthalmometer is now at zero. Then, by turning the screws already mentioned, the glass plates in the interior of the box are placed obliquely, and the motion is continued until six small specks of light are seen thus :—

*	*	*	*	*	*
1	*a*	2	*b*	3	*c*

Here the asterisks numbered 1, 2, 3, represent one-half of the amount of displacement in the one direction, and those marked *a*, *b*, and *c*, half the displacement in the other direction. Thus the three original images have been displaced through a distance equal to that between the two extreme images. The number of degrees through which the plates have moved are now read off, giving the angle of incidence, and by means of the calculation above described, the size of the image is ascertained. The following measurements are now made : First, the distance between the upper and lower reflecting mirrors—this gives the size of the object ; and second, by a tape line divided into millimetres, the distance from the anterior surface of the cornea to the centre of the apparatus bearing the mirrors ; but as these reflect rays of light from the candle flame, this measurement must be doubled. We now know the size of the object, the size of the image, and the distance of the object from the cornea ; and from these data, by the formula already given,

the radius of curvature is easily calculated. (For an example, see p. 565.)

Donder's method of using the ophthalmometer.—This is an easier, though a less accurate, mode of measuring reflected images. It consists of preparing a scale, in which each degree, or fraction of a degree, corresponds to a certain size in millimetres. The mode of constructing it is as follows: Place a small white ivory scale, divided into tenths of a millimetre, at a distance of ten feet from the ophthalmometer. Turn the plates until the lines on the scale diverge and ultimately pass through any given distance, say $\frac{1}{10}$ of a millimeter, $\frac{1}{8}$, $\frac{1}{6}$, $\frac{1}{4}$, $\frac{1}{3}$, $\frac{1}{2}$, $\frac{3}{4}$, or 1 millimeter. The number of degrees corresponding to each of these displacements is noted, and thus a scale, by numerous observations, may be readily constructed. Observations are to be made on the living eye as described. When the distance between two reflections on the cornea, representing the image, has been displaced through its whole extent, the number of degrees are noted, and on referring to the scale of measurements prepared as above, the size of the image is at once known.

Measurements made by means of the ophthalmometer.—We give here various measurements in millimetres made by means of the ophthalmometer, which are intended to serve as standards of comparison for students or others who may make a special study of, and devote time to, the subject :*

Optical Constants.	Myopic eye. *Knapp.*		Hypermetropic eye. *Woinow.*		Presbyopic eye. *Adamück and Woinow.*	
	Rest.	Accommoda'n.	Rest.	Accommoda'n.	Rest.	Accommoda'n.
Radius of cornea . .	7·2053	7·2053	8·00747	8·00747	7·15568	7·15568
Radius of anterior surface of lens . . .	9·0641	5·0296	9·3785	5·2904	10·2021	8·5975
Radius of posterior surface of lens . .	6·4988	5·0855	6·2480	4·9714	6·2156	5·0001
Distance of anterior surface of lens from the cornea . . .	3·4786	2·8432	3·6175	3·0028	3·23731	·2·98985
Thickness of the lens .	3·6225	4·2579	3·5825	4·1972	[3·96269	4·21015
Focal distance of the lens	43·133	30·939	44·9616	31·185	46·357	38·1513
Index of refraction of the cornea, aqueous humour and vitreous humour .	1·3465†	...	...	...	...	...
Index of refraction of the lens	1·4545†	...	...	...	...	...

* M. Woinow, Ophthalmometrie, Wien. 1871. † Helmholtz, Physiologische Optik.

*The apparatus of Knapp for holding the head.**—This apparatus, designed by Professor Knapp, now of New York, is for the purpose of securely holding the head of the individual whose eye is being examined with the ophthalmometer. It is composed of a circular piece of wood placed vertically, which may be turned round on an axis, and which has holes cut for the nose, eyes, and mouth. The head is held steady by two flat pieces of wood, which are moved by screws so as to be applied, one to each side of the head. These are well padded.

The phakoscope of Helmholtz (φακη, the lens; σκοπεω).—This is an irregularly triangular box, shaped somewhat like a stereoscope, made of wood or paste-board, and well blackened in the interior. It has four apertures, one for admitting the rays of light from a candle or lamp, another for the eye to be examined, and a third for the eye of the observer. The fourth opening is immediately in front of that for the eye to be examined, and is furnished with a sliding door bearing a small vertical needle. By causing the person whose eye is being examined to direct his attention to the needle, the eye is accommodated for near distances, but if the needle be removed by pushing down the little door, the eye may accommodate itself to any distant object. When an eye is examined in this manner three reflections are seen, one from the anterior surface of the cornea, Fig. 24^a 1, a second from the anterior surface of the lens, 2, and a third, which is inverted, from the posterior surface of the crystalline lens or anterior layer of the vitreous humour, 3. The relative positions of these images, when the eye is accommodated for distant objects, is seen in Plate XXI. fig. 24^a, while that represented in Fig. 24^b shews the position in which they are in an eye looking at a near object. It will be observed that in the latter, the anterior surface of the cornea has become more convex, because the image 2 in Fig. 24^b has advanced nearer to 1 than in Fig. 24^a. This is the most suitable instrument for demonstrating the accommodation of the eye to distance according to Cramer (see p. 342).

The ophthalmoscope.—This valuable instrument, also invented by Helmholtz, is for the purpose of illuminating the posterior chamber of the eye, so as to enable us to see the retina, the entrance of the optic nerve, &c. There are many varieties

* Knapp, Archiv. v. Ophthalmol. Bd. VI. Ab. 2, and Archiv. f. Augen und Ohrenheilk, 1 Bd. 2 Ab.

of ophthalmoscopes, which it is unnecessary to describe here ; but they all consist essentially of a powerful mirror, having a small round hole in its centre, which is used for reflecting a beam of light into the eye under examination ; and of a biconvex lens for the purpose of magnifying the retina, and of projecting an image of the latter forwards to a point somewhere between the eye of the demonstrator and the eye of the patient. The mode of using the instrument is as follows : Let the individual whose eye is to be examined, be seated in a dark room on a chair of a convenient height, and place a gas light, furnished with a ground-glass shade, or a moderator lamp, immediately behind his shoulder, and yet close to the side of his head. Let the light be on a level with the eye, and so far behind it that the countenance is in the shade. If this cannot be so arranged, place a dark shade between the eye and the light. The student is now to sit down opposite, take the mirror in his right hand and apply the back of it to his own eye, at a distance of about eighteen inches from the eye under examination, and looking carefully through the small hole in the back of the mirror, he so moves the latter as to catch the rays of the light from the lamp, and to reflect them into the patient's eye. He must now take the biconvex lens between the forefinger and thumb of the left hand, and hold it at a distance of about an inch in front of the patient's eye, keeping the hand steady by resting the little finger on the forehead of the patient. Then by a slight backward and forward movement of his head and the mirror, the student will succeed in catching the proper focal distance, and the image of the retina will then be distinctly seen.

In the axis of the eye-ball the *yellow spot* (*macula lutea*) may be seen forming a slight oval patch, and having in its centre a small bright dot, the *fovea centralis*, the thinnest part of retina. About the 1-10th of an inch to the inside of the yellow spot, we now observe a white or rose-coloured round spot, bounded by a well-defined border, and having radiating from its centre a number of minute vessels. This is the entrance of the optic nerve, and the vessels are branches of the central artery of the retina. It is sometimes termed the optic disc (*porus opticus*), and it has also been called the optic papilla, because at this place the nervous substance of the retina is slightly elevated, so as to form an eminence (*colliculus nervi optici*). The neighbourhood

of the optic disc and yellow spot is usually of an orange-red colour, and is richly supplied with branches of the retinal vessels. The yellow spot is seen most distinctly when the individual looks straight forward, and the optic disc is demonstrated when the eye is rolled a little inwards. By causing the patient to move his eye in different directions, the whole surface of the retina may be examined.

Experiments on Hearing.

This department of practical physiology has become, chiefly owing to the brilliant researches of Helmholtz, one of the most inviting fields of study. Before entering upon it, however, the student should be familiar with the general principles of acoustics explained at pp. 129, 134. The following are a few examples of the kind of apparatus used, and the experiments performed.

Helmholtz's model for explaining the mechanism of the bones of the ear.—This is a model of the tympanum, having accurate models of the *malleus*, *incus*, and *stapes*, moving upon each other by joints, and having attached to them, at the proper points, cords which run in the direction of, and represent the muscles of, the tympanum, viz., the *tensor tympani*, *laxator tympani*, and *stapedius*. By tightening or relaxing these cords, the action of the chain of bones or the *membrana tympani*, on the one hand, and on the membrane covering the *fenestra ovalis*, on the other, may be easily demonstrated. It may also be shewn by means of this apparatus how the vibrations of the air communicated to the *membrana tympani* are carried onward through the chain of small bones to the *fenestra ovalis*.

Monochord of Helmholtz.—This is an apparatus consisting of a long narrow box, on the upper surface of the lid of which there is a cord drawn tightly over two ivory bridges, placed one at each end of the box. There is also a bridge, of the shape of that of a violin, which may be moved in any direction beneath the cord; and thus it may be shewn that vibrating cords of different lengths produce various musical notes. Attached to the instrument there is a trumpet-shaped resonating apparatus, having the wide end covered with a delicate membrane. This part of the apparatus may be moved along the side of the box, so as to be opposite to any given point of the vibrating

cord, and the note corresponding to the vibrations will be distinctly heard.

The apparatus of Appunn for illustrating the researches of Helmholtz, adapted for physiological purposes.—This is an apparatus invented and made by Georg Appunn, of Hanau, near Frankfort-on-the-Maine.* It consists of a strong table provided with a bellows and air-chest. The entrance of air from the bellows into the chest is regulated by a valve, which may be opened or shut at pleasure. On the top of the table there are six square holes communicating with the air-chest, each being provided with a sliding valve, which can be opened or shut by a rod attached to it. These square holes serve as sockets, into which we can fit the different parts of the apparatus. When we wish to use any particular instrument, the valve beneath it is opened, and thus air is at once admitted into it from the air-chest.

The following are the principal parts of the apparatus :—

1. *An over-tone apparatus.*—This part of the instrument consists of a rectangular chest, having in its interior sixty-four metallic tongues, placed transversely, and carefully constructed, so that the vibration of each produces a certain musical tone. Suppose we were to divide a vibrating cord into equal parts of $\frac{1}{2}$, $\frac{1}{3}$, $\frac{1}{4}$, $\frac{1}{5}$, $\frac{1}{6}$, and so on up to the 1-64th of its length, we would obtain the tones of this apparatus. Underneath each tongue there is a sliding stop, which has a short metallic rod attached to it. The end of each rod is finished with a small knob, by pulling out which, the sliding valve under any particular tongue may be withdrawn, and the corresponding musical tone produced by working the bellows. In order to maintain the sound while the bellows is refilling, the top of the box of the over-tone apparatus moves upwards and downwards like an accordion, and the tongues vibrate whether the air passes from below upwards, or from above downwards. By means of this apparatus, as we can sound several notes at the same moment, the peculiar colour, or timbre, or, according to Helmholtz the *klang* of a musical note may be demonstrated. Every musical note contains not only its fundamental tone, and overtones, but also other tones called the octave, the duo-decimo, double octave, &c., the ratio of the vibrations of which are as 1 : 2 : 3 : 4 : 5 : 6 : 7, and so on. In order to detect these over-

* Ueber die Helmholtz' sche Lehre von den Tonempfindungen, &c.

tones, we require a series of instruments termed *resonators*. These are globes or cones made of copper or tin, and each one is so constructed that the air in its interior is thrown into vibration by one of the over-tones in the musical note. With Appunn's apparatus, twenty-nine of these resonators are supplied. They vary in length from $4\frac{1}{2}$ feet to 3 inches, and they are all conical and made of tin. The narrow end is placed close to the ear, and the base directed upwards, and if possible the observation should be made at a distance from the place or instrument from which the sound issues. The presence of over-tones in a musical note may be shewn by sounding the fundamental tone. Then, by withdrawing the stops corresponding to the different over-tones of that particular note, we find that its colour or quality depends on the different number and relation of the over-tones. It is this fact which explains why the same note on a flute, a clarionet, a piano, or a trumpet, differ from each other in quality. The fundamental tone is the same in each, but the number and relation of the over-tones vary. The soft note of a flute contains fewer over-tones than the same note sounded on a trumpet. For instance, if we sound on the over-tone apparatus the notes in the proportions 1 : 2 : 3 : 4 : 5 : 6 : 7, &c., which are the over-tones of the fundamental tone = 1, the sound becomes stronger and rougher as we proceed. This illustrates the law laid down by Helmholtz, that "The more over-tones a compound note contains, the rougher is its quality or timbre." The quality of the voice in different individuals depends on the number and strength of these over-tones.

Another method of observing the over-tones is to sound the fundamental note on the over-tone apparatus, and the students may hear distinctly the various over-tones corresponding to the particular note by listening with the resonators at the other end of the room. In order to sound the fundamental note with volume and intensity, so that the over-tones may be detected in a room with a large audience, Appunn's apparatus is provided with two powerful *tongue-pipes*. These consist of rectangular wooden pipes, having at the top a wedge-shaped box, the apex of the wedge pointing downwards, and furnished with a vibrating metallic tongue. A large cone made of tin is fitted, apex downwards, into the top of the pipe. This acts as a powerful

resonator, and the note produced by the metallic tongue is increased in intensity.

After some practice with this instrument, one can detect the over-tones even without the resonators. The resonators do not produce the over-tones, but the confined mass of air within them, by its vibrations in unison with those causing the over-tones, strengthens the latter, and renders them appreciable to the human ear. The over-tones are not therefore formed in the ear, as was supposed before the researches of Helmholtz, but they are produced in the air surrounding the apparatus causing the fundamental note.

The production of combination tones.—If, by the over-tone apparatus, we sound two notes of different pitch at the same time, and if they correspond in strength, we may hear, by using a resonator, not only the two primary notes, but a third, which is deeper than the primary. This is termed the combination or ground-tone, first discovered by Andreas Sorge in 1740, and investigated by a violinist, Tartini, and often called after the latter, *Tartinian tones.* For instance, if we sound two notes on the over-tone apparatus, the proportion of whose vibrations are as 2 : 3, or 3 : 4, or 4 : 5, or 6 : 7, or 7 : 8, we hear the ground tone, which is always, in this case, the tone whose vibrations are as $1 = C_2$. The student, until his ear is accustomed to the apparatus, should sound the notes 16 : 20, or 20 : 24, or 24 : 28, and he will easily hear the deep ground tone, the vibrations of which will stand in the arithmetical ratio of 4 to the figures just given. In the same way the proportions 16 : 18 : 20 : 22 : 24 produce a ground-tone = 2; those of 15 : 18 : 21 : 24 : 27 produce tone = 3; and so on.

The production of difference tones.—The difference tone is that in which the number of its vibrations are equal to the difference between those of the two primary tones. For example, suppose we sound on the over-tone apparatus two notes, the ratio of whose vibrations are as 16 : 20. The difference tone will have a vibration of 4, that is, 20 — 16 = 4. But the vibrations of 1 (the lowest in the over-tone apparatus) = 32 in the second. Therefore, the vibrations of 16 will be $16 \times 32 = 512$; of 20 will be $20 \times 32 = 640$; and of the difference tone will be $4 \times 32 = 128$ vibrations in an equal period of time. This tone was first discovered by Helmholtz.

The production of summation tones.—When we sound two

tones of unequal pitch, but the ratio of the vibrations being as 16 : 18 : 20 : 22 : 24, &c., we obtain not only, as already described, a fundamental or ground tone, and a difference tone, but we also produce a third called a summation tone. For example, if we sound on the over-tone apparatus the tones 4 : 6, we hear, by means of resonator No. 10, another tone, the vibrations of which are in the proportion of 10 : 4 : 6. We hear in those circumstances a musical chord, thus (e = summation tone):

C : G : e
4 : 6 : 10

And in the same way by sounding 1 : 2, 2 : 3, 3 : 4, or 4 : 5, we may produce the corresponding summation tones, 3, 5, 7, 9, &c.; and these summation tones may be heard with resonators 3, 5, 7, 9, &c.

By this apparatus also, even higher tones may be obtained. Thus, when we sound 2 : 3, we may hear the deep difference tone = 1; at the same time the summation tone $2 + 3 = 5$; and with care, by increased attention, we may hear another tone, the ratios of whose vibrations are as $2 \times 2 + 3 = 7$; and even a third, namely, that produced by $2 \times 2 + 2 \times 3 = 10$. Thus, when we sound 2 : 3, and provide four individuals with the resonators marked 1, 5, 7, and 10, each will hear distinctly the tone corresponding to the resonator he employs.

2. *An apparatus to demonstrate deep difference tones.*—This consists of two tall organ pipes placed at the end of the table, and communicating with the air chest. These pipes are of equal length, but each is furnished with a stop attached to a vertical rod, by which the length of the vibrating column of air in the one may be made to differ from that in the other. When the stops are so arranged that the columns of air in the two pipes are equal, and the bellows are worked, we obtain two notes so perfectly in unison as to be undistinguishable. If, however, the length of one of the columns of air be diminished by pushing down the stop, the rate of vibration is changed in that pipe, and we hear two notes or impulses alternating with each other; a kind of shake. It may also be shewn that these impulses increase in rapidity as we augment the difference between the lengths of the columns of air in the two pipes, that is as the vibrations in the two differ. And it may be demonstrated that the number of impulses per second is always equal to the difference between the rates of vibration.

3. *A Vocal apparatus.*—This will be described with the experiments on voice (see p. 579).

4. *Appunn's tone-measurer.*—This consists of an apparatus similar in structure and appearance to the over-tone apparatus already described. It contains thirty-three tones, marked 0, 1, 2, 3, 4, 5, &c., on to 32; and these tones are so arranged that the difference between the vibrations of any adjacent two, such as 0 : 1, or 1 : 2, or 2 : 3, and so on, produce, when sounded together, exactly *four* impulses per second. In the same way when 0 : 2, or 1 : 3, or 2 : 4 are sounded, we have eight impulses per second; again 0 : 3, or 1 : 4, or 2 : 5 give sixteen impulses per second, and so on. With this instrument we can readily determine the number of vibrations in any given tone (see p. 347).

Experiments on Voice.

The human voice is produced by the vibrations of the true vocal cords (p. 351). This may be illustrated in several ways.

1. *By Müller's artificial larynx.*—This consists of a wooden tube of the form seen in Plate XXI. fig. 25, *f*, having at *e* a brass frame-work for holding a piece of india-rubber tube, and moving on a hinge, so that the margins of the latter may be separated or approximated, or relaxed or rendered tense. On blowing through the end of the tube, various sounds are produced according to the degree of tension of the india-rubber margins. It may be shewn that as we increase the tension of these, the pitch of the sound rises, while it is lowered when we remove the tension.

2. *By Müller's apparatus for experimenting on the vocal cords themselves.*—A view of the principal portion of this apparatus is seen in Plate XXI. fig. 25. It consists of a broad wooden stand, having two or three strong uprights, provided with numerous holes and screws, by which the larynx may be attached. In Fig. 25, the anterior part of a human head, with the larynx left attached to it, is affixed to the apparatus. Into the trachea there is inserted and securely fastened the bent wooden tube *f*, by which air can be forcibly blown upwards through the glottis. When this is done, a sound is produced, which may be increased in pitch in two ways: 1st, by means of the rectangular forceps *a*, movable on the steel rod *b*, which grasp the larynx and thus approximate the cords; or 2nd, by fixing a small hook into the anterior and upper border of the thyroid cartilage, and

carrying a thread from this hook over the pulley *c*. By placing weights in the scale or cup *b d* attached to the thread, the thyroid cartilage is pulled forwards and the cords tightened.

Instead of using part of a head, as shewn in Fig. 25, it is more satisfactory to make a special preparation of a larnyx, by removing it from the neck, clearing it from the muscles, and by cutting off the upper part down to the level of the true vocal cords. It may then be clearly demonstrated that voice is produced by the vibrations of these cords, and numerous experiments may be performed, shewing how they are influenced by the various muscles, and the change of note consequent thereon.

Formation of vowel sounds.—These may be illustrated by the special apparatus made by Appunn, already alluded to. It consists of a combination of wooden organ pipes, having stops so that air may be admitted into one or more at pleasure. It may be shewn that the vowel *a* consists of a fundamental note and of certain over-tones; the vowel *e* of a fundamental note and ther over-tones, and so on. The fundamental note is produced by the vibrations of the cords, and is the same for all the vowels; but the over-tones are produced in the upper part of the larnyx, and are modified by the resonance of the mouth. It is well known that the vowels, if sounded by different voices, have not the same quality or timbre ; a fact due partly to the over-tones varying in different individuals, and partly to the effect of resonance of the oral cavity, which also varies in shape. (See p. 353.)

3. *The Laryngoscope.*—The idea of illuminating and rendering the larynx visible by means of a reflector, has been more or less attempted by Liston, Warden, Avery, Garcia, and others, but abandoned as impracticable in medicine, until successfully revived in recent times (1858–59) by Professor Czermak. For the examination of the larynx he employs, 1st, a perforated mirror, by means of which a powerful light is thrown from a lamp into the back of the mouth, and through which the operator gazes in the direct axis of the illuminating rays. This mirror may be attached to a bent stalk, the end of which can be held firmly by the teeth, but it is far more conveniently, for purposes of demonstration, held firmly in the left hand. 2d, A laryngeal mirror of glass or steel, varying in size, attached to a stem at one of its corners, which having been previously warmed to pre-

vent condensation of the breath upon it, is placed in front of the uvula, and reflects the image of the rima glottidis to the eye of the observer.

The person examined should place his hands upon his knees, the upper part of the body is advanced forwards, the neck bent onwards, the nape slightly inclined backwards, the mouth widely open, the tongue flattened and held a little without. The observer is seated in front of the person to be examined; he places in his mouth the handle which supports the illuminating mirror, and looks through the central opening; the laryngeal mirror, introduced into the back part of the mouth with the right hand, is illuminated by the light which is projected from the illuminating mirror. In the first place, the illumination of the back part of the mouth and the mutual position are regulated; then the laryngoscope is heated, and its temperature regulated by the touch. After these preliminaries are gone through, the patient should open the mouth wide, and alternately inspire and expire deeply. While doing so, the laryngoscope is placed against the uvula and the *velum palati*, to sustain these parts a little, and the mirror is given a convenient inclination; at times it is impossible to avoid touching the posterior wall of the pharynx; the examination is directed by the image we thus obtain. On telling the patient to pronounce ah! the movement of the vocal cords is seen. Practice and reflection will bring each observer to comprehend the modifications to which he ought to submit this proceeding, according to the special circumstance; whether, for instance, he has in some degree to advance or to withdraw the laryngoscope, to bend it, to lower or to elevate it, to change the position and attitude of the individual undergoing examination, raise his chair.

For the performance of many experiments, more especially those on the nervous centres and individual nerves, anatomical knowledge and operative skill are required. To these we have not specially alluded, neither do we profess to have exhausted those for which particular instruments have been invented; but enough has been said to illustrate this department of physiology, and to assist the student in making himself familiar, practically, with the use of the extensive apparatus he will find in the laboratory.

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ERRATA.

Page 2, line 13 from bottom, *for* "C_2 N," *read* C N.
" 6, " 18 from top, *for* "of the above series," *read* of the first of the above series.
" 6, " 11 from bottom, *omit* "free."
" 10, " 4 from bottom, *for* "Ptalyin," *read* Ptyalin.
" 14, " 15 from top, in formula for urea, *for* "O_2," *read* O.
" 16, " 1 at top, in formula for xanthin, *for* "O," *read* O_2.
" 16, " 16 from bottom, in formula for cholic acid, *for* "H_{49}," *read* H_{40}.
" 28, " 3 from top, *for* "latter," *read* former.
" 28, " 13 from bottom, *for* "Ca Fl," *read* Ca F_2.
" 29, " 10 from bottom, *for* "possible," *read* impossible.
" 37, " 17 from bottom, *for* "rises," *read* rise.
" 38, " 9 from top, *for* "a'l," *read* cell.
" 63, " 2 from top, *for* "but no fats," *read* and phosphuretted fats.
" 63, " 5 from top, *for* "renders," *read* render.
" 64, " 6 from bottom, *for* "some," *read* semi.
" 74, " 1 at top, *for* "consists," *read* consist.
" 77, " 14 from top, *omit* "a," and *for* "body," *read* bodies.
" 80, " 21 from bottom, *for* "have" and "exist," *read* has and exists.
" 81, " 21 from top, *omit* "basic."
" 83, " 7 from bottom, *for* "commences," *read* commence.
" 90, " 4 from top, *for* "consists," *read* consist.
" 112, " 19 from bottom, *for* "straight," *read* straight or curved.
" 121, " 14 from top, after "gas," *introduce* at zero C.
" 125, " 9 from bottom, after "volume," *introduce* at zero C.
" 131, " 1 at bottom, *for* 27, 27, *read* 24, 27.
" 138, " 15 from bottom, *for* "Fraunhofer," *read* Frauenhofer.
" 146, " 14 from bottom, *for* "magnetic," *read* diamagnetic.
" 147, " 2 from top, *for* "inherent," *read* not inherent; and *for* "and evolved from them," *read* but developed in them.
" 147, " 10 from top, after "is," *introduce* in.
" 147, " 25 from top, *for* "intervals," *read* interval.
" 152, " 10 from bottom, *for* "Zn SO_4," *read* Cu SO_4.
" 155. " 15 from bottom, *for* "form," *read* kind.
" 158, " 8 from top, *for* "was," *read* were.
" 164, " 17 from bottom, *for* "fig. 24," *read* fig. 34.
" 172, " 23 from top, *for* "opened," *read* closed.
" 172, " 24 from top, *for* "closed," *read* opened.
" 172, " 7 from bottom, *for* "clos." *read* open.
" 175, " 17 from top, before "nerve," *introduce* the.
" 178, " 3 from bottom, *for* "are," *read* is.
" 179, " 17 from bottom, *for* "sensations," *read* secretions.
" 182, " 7 from bottom, *omit* "gravity."
" 183, " 15 from top, *for* "his," *read* its.
" 185, " 17 from bottom, *for* "produce," *read* produces.
" 186, " 12 from top, after "which," *introduce* its.
" 190, " 17 from bottom, *for* "consists," *read* consist.
" 193, " 1 at top, *for* "latter," *read* former.
" 193, " 2 from top, *for* "former," *read* latter.
" 194, " 13 from top, *for* "is," *read* are.

Page 199, line 5 from top, *for* "action," *read* section.
,, 200, ,, 16 from top, *for* "are," *read* is.
,, 202, ,, 9 from top, *omit* "of Peyer and."
,, 215, ,, 2 from bottom, *for* "Poissieulle," *read* Poisseuille.
,, 216, ,, 19 from top, *for* "is increased," *read* are increased.
,, 219, ,, 17 from top, *for* "Hæmadrometer," *read* Hæmadromometer.
,, 224, ,, 9 from top, *for* "possess," *read* possesses.
,, 224, ,, 2 from bottom, *for* Plate IX.," *read* Plate XI.
,, 227, ,, 16 from top, *for* "by," *read* of.
,, 233, ,, 12 from top, *for* "selenurretted," *read* seleniuretted.
,, 234, ,, 11 from top, *for* "function," *read* functions.
,, 240, ,, 4 from top, *for* "some," *read* same.
,, 241, ,, 17 from bottom, *for* "liquid," *read* liquids.
,, 242, ,, 3 from top, after "these," *introduce* latter.
,, 247, ,, 3 from bottom, *for* "Helmholz," *read* Helmholtz.
,, 249, ,, 21 from top, *for* "contain," *read* contains.
,, 255, ,, 10 and 12 from top, *for* "kilos," *read* grammes.
,, 255, ,, 5 from bottom, *for* "shew," *read* shews.
,, 256, ,, 22 from top, *for* "corticle," *read* cortical.
,, 256, ,, 8 from bottom, *for* "uretors," *read* ureters.
,, 258, ,, 8 from bottom, *for* "32," read 62.
,, 260, ,, 7 from bottom, *for* "anasarea," *read* anasarca.
,, 262, ,, 9, 12, and 13 from bottom, *for* "divided," *read* derived.
,, 263, ,, 6 from bottom, *for* "chorium," *read* corium.
,, 273, ,, 7 from top, *for* "absorption or decomposition," *read* decomposition and absorption.
,, 273, ,, 2 from bottom, *for* "undergo," *read* undergoes.
,, 274, ,, 4 from bottom, *for* "seven," *read* fifteen.
,, 276, ,, 6 from top, after "to," *introduce* help to.
,, 318, ,, 16 from bottom, after "convulsed," *introduce* and the eye is turned outwards.
,, 318, ,, 14 from bottom, *for* "outwards," *read* inwards.
,, 329, ,, 9 from bottom, *for* "Eckar," *read* Ecker.
,, 338, ,, 13 from top, *for* "opthalmotrope," *read* ophthalmotrope.
,, 342, ,, 11 from bottom, *for* "opthalmometer," *read* ophthalmometer.
,, 344, ,, 22 from bottom, *for* "right," read left.
,, 352, ,, 10 from top, *for* "differ," *read* differs.
,, 352, ,, 8 from bottom, *for* "precision to," *read* precision from.
,, 360, ,, 4 from top, *for* "contain," read contains.
,, 361, ,, 4 from bottom, *for* "preversions," *read* perversions.
,, 380, ,, 13 from bottom, *for* "(Fig. 3, *h* to *k*)," *read* (Fig. 3, *m n o*).
,, 380, ,, 10 from bottom, *for* "(Fig. 3, *m* to *r*), *read* (Fig 3, *p q* and *h* to *k*).
,, 387, ,, 2 from top, *for* "vertebræ," *read* vertebra.
,, 389, ,, 8 from bottom, after "circle" *omit* the.
,, 390, ,, 10 from top, *for* "cerebral," *read* central.
,, 394, ,, 17 from bottom, *for* "mesteric," *read* mesenteric.
,, 395, ,, 9 from top, after "passing," *introduce* from.
,, 395, ,, 14 from bottom, *for* "thryroid," *read* thyroid.
,, 403, ,, 16 from bottom, *for* "tasselated," *read* tesselated.
,, 452, ,, 9 and 10 from top, *for* "Guiacum," *read* Guaiacum.
,, 501, ,, 16 from bottom, *for* "acromatic," *read* achromatic.
,, 549, ,, from top, Greek character is inverted.

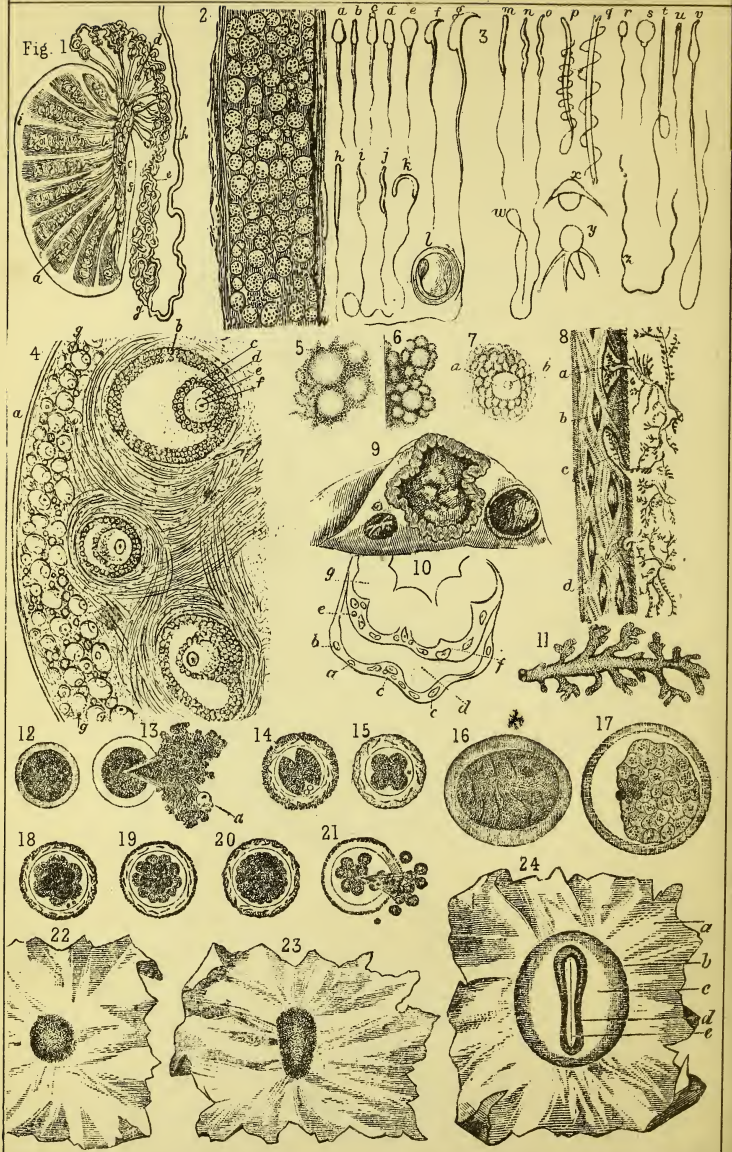

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Plate XVII.—Reproduction.

Fig. 1. Section of a testicle. *a.* Convoluted *tubuli seminiferi; b, s. vasa recta; c, f. rete testis,* in the *corpus Highmorianum; d. globus major; e.* body of the epididymis; *g. globus minor; h. vas deferens; i i. tunica albuginea.*

Fig. 2. View of a portion of one of the *tubuli seminiferi,* shewing molecular matter, and cells of various sizes in which the spermatozoids are developed. (250 *diam.*)

Fig. 3. Spermatozoids of various animals. *a.* Spermatozoids of man, viewed on the surface; *b.* the same viewed edgeways; *c.* the same with granule at summit of head; *d.* the same edgeways; *e.* dog; *f.* mouse; *g.* rat; *h.* frog; *i.* snake; *k.* lizard; *l.* spermatozoid coiled up in cell; *m.* wild duck; *n.* shrike; *o.* finch; *p.* Bombinator igneus; *q.* a magnified view of a portion of the same; *r. perch; s.* loach; *t.* shark; *u.* lamprey; *v.* Helix (a snail); *w.* planaria; *x.* a crab; *y.* Pagurus (hermit crab); *z.* earthworm.

Fig. 4. Section of a portion of an ovary. *a.* Fibrous coat; *b.* a Graafian vesicle, shewing at c the *tunica granulosa; b. membrana granulosa; d. zona pellucida* and yolk; *e.* germinal vesicle; *f.* germinal spot; *g.* Graafian vesicles forming. In the middle of the figure, a Graafian vesicle is seen in a more advanced stage of development, and at the bottom we have one still farther advanced. These lie in a fibrous stroma.

Figs. 5 and 6. Formation of ova by molecular aggregation.

Fig. 7. A primitive Graafian vesicle, *a,* containing an ovum *b.*

Fig. 8. Section of the mucous membrane of the uterus, shewing at *a, c,* and *d,* a fœtal tuft projecting into a uterine sinus: and at *b,* another sinus.

Fig. 9. Section of an ovary shewing in the centre a recent *corpus luteum,* on the right hand an older one, and on the left one still older, much reduced in size by contraction.

Fig. 10. The extremity of a placental villus. *a.* The external membrane of the villus,—the lining membrane of the vascular system of the mother; *b.* the external cells of the villus,—cells of the central portion of the placental decidua; *c, c.* germinal centres of the external cells; *d.* the space between the maternal and fœtal portions of the villus; *e.* the internal membrane of the villus,—the external membrane of the chorion; *f.* the internal cells of the villus, the cells of the chorion; *g.* the loop of umbilical vessels.

Fig. 11. A placental villus.

Fig. 12. An ovum from the bitch freed from the granular membrane, shewing the dark internal yolk, and clear external *zona pellucida.* (50 *diam.*)

Fig. 13. The same ovum lacerated with a needle. The yolk has flowed out, shewing the germinal vesicle, *a,* with its germinal spot. (50 *diam.*)

Fig. 14. The ovum has encountered spermatzoids, which are seen adherent to the *zona pellucida.* Fecundation has taken place; the spermatzoid, which has penetrated the transparent zone, together with the germinal vesicle, has been dissolved in the yolk, which is divided into two masses. (50 *diam.*)

Fig. 15. The yolk divided into four masses. (50 *diam.*)

Figs. 18 and 19. The process of division in the yolk further illustrated. (50 *diam.*)

Fig. 20. The yolk now reduced by division to a large number of molecular cells. (50 *diam.*)

Fig. 21. The molecular cells rendered visible by laceration of the ovum. They contain a clear space in their centres. (50 *diam.*)

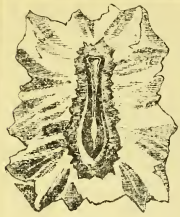
Fig. 17. An ovum further developed after it has been placed in water for a short time. In consequence of endosmose, the internal membrane is separated from the *zona pellucida,* and is seen to be formed by the cells which have coalesced. This is the germinal membrane with the germinal area composed of an extra layer of cells. (50 *diam.*)

Fig. 16. An ovum much larger, taken from the uterus, moistened with water. The germinal membrane is somewhat separated from the *zona pellucida,* and thrown into folds. (10 *diam.*)

Fig. 22. Portion of the germinal membrane surrounding the germinal area, cut out from a further developed ovum. A clear space in the area called *area pellucida* is apparent. (10 *diam.*)

Fig. 23. A similar piece from a somewhat older ovum. The germinal area has become oval. (10 *diam.*)

Fig. 24. The germinal area is now greatly enlarged in the germinal membrane; *a.* germinal membrane; *b.* limit of vascular area; *c. area pellucida; d. laminæ dorsales; e.* Primitive groove.



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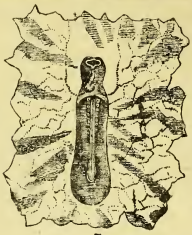

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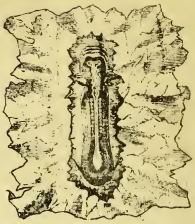

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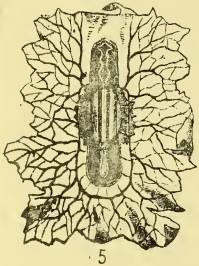

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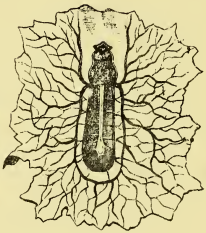

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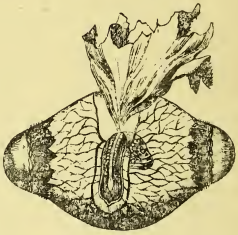

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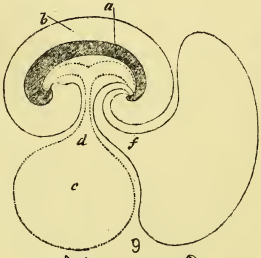

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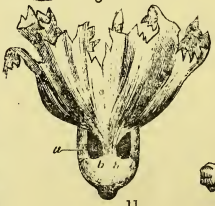

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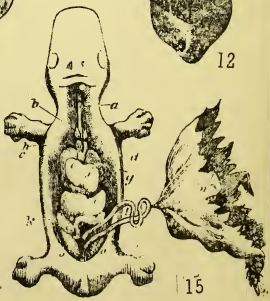

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Plate XVIII.—Reproduction.

Fig. 1. Portion of germinal membrane, with the embryo from an ovum twenty-four hours older than Fig. 24, Plate XVII. The primitive groove is not yet closed, but is much stronger, especially above. Here three swellings are observable, which are the three primitive brain-cells. At the inferior end, the groove is of a lancet shape (*sinus rhomboidalis*). In the centre of the groove is a thin streak, the commencement of the *chorda dorsalis*. Six square cells are formed on each side, the commencement of the vertebral column. The germinal membrane is now composed distinctly of two layers, the upper of which (*the serous* or *animal layer*) is cut close round the embryo, shewing more distinctly the lower (the *mucous* or *vegetative layer*). (10 *diam.*)

Fig. 2. The same embryo, seen sideways, whereby the elevation of the dorsal laminæ, and the groove between them, are better seen. The head is already distinctly elevated above the germinal membrane. (10 *diam.*)

Fig. 3. An embryo twelve hours older than the former one, turned round and examined on the under or abdominal surface. The head with the broadened-out first brain-cell is seen coming forward. Immediately below this an S-shaped tube is seen, which is the rudimentary heart. The lower end branches off on each side to join the vascular network, forming the *venæ omphalo-mesentericæ*. The visceral or abdominal cavity is seen open below, causing the embryo to resemble somewhat the appearance of a partly-decked boat. (10 *diam.*)

Fig. 4. The same embryo seen from above. The primitive groove is now for the most part closed over. The first brain-cell is widened out laterally, and bent forwards. The posterior ones are altered in shape from absorption of fluid. There are two vertebral cells. At both ends of the primitive groove folds of the serous layer are visible—the commencement of the amnios. The serous layer is cut close round the embryo ; and upon the mucous layer, fine lines, in the form of a network, are visible—the commencement of the *vascular layer*. (10 *diam.*)

Fig. 5. An embryo from an ovum supposed to be twenty-three or twenty-four days old, seen from above. The primitive groove is now completely closed, to form the medullary tube, and exhibits above the three primitive brain-cells. The first of these is seen to be so expanded laterally as to form at each side the embryo eyes. The embryo ears are also seen at each side opposite the third brain-cell. The upper and lower ends of the embryo are now inclosed in a backward fold of the serous layer, which, however, is still open in the centre. The blood vessels in the vascular layer are now fully formed. (10 *diam.*)

Fig. 6. The same embryo seen from below. The head is strongly bent forward, so that the first brain-cell and embryo eyes are best seen on this surface. Below these, two notched processes are seen, which are the first visceral arches. Below these again, the S-shaped heart—terminating, above in the aorta, below in the *venæ omphalo-mesentericæ*. The heart now pulsates, and a circulation is established over the vascular area. (10 *diam.*)

Fig. 7. An entire ovum, with the embryo somewhat older than the last. The villous chorion is raised off the entire centre of the egg, which is suspended by it at one point, where the folds of the serous layer have completely closed over the back to form the amnios. The embryo lies with its interior half in the plane of the vascular and mucous layers ; whilst the head and superior half is prominent, and inclosed by them. At the sides are seen both the *arteriæ* and *venæ omphalo-mesentericæ*, which communicate with the plexus of the vascular layer, and terminate in circular rings, the *venæ terminales*, leaving the two poles of the ovum bare. (5 *diam.*)

Fig. 8. The same embryo, removed with its membranes, and viewed from the internal surface of the ovum, sideways. The head and upper portion is seen surrounded by the amnios. In the head is observable the brain, divided into anterior, neighbouring, and middle brain, *a*, *b*, *c* ; the third brain-cell, *d* ; eyes, *e* ; ears, *f* ; not yet connected with the third brain-cell. There are three visceral arches. The heart is further developed, prominent, and sur-

rounded by the serous membrane. The lower portion of the embryo is covered with the vascular and mucous layers. (5 *diam.*)

Fig. 9. Diagram representing the mode of formation and position of the three embryonal sacs. *a*, Embryo—*b*, amnios—*c*, umbilical vesicle—*d*, the vitelline duct, or pedicle of the umbilical vesicle—*e*, allantois—*f*, the urachus or pedicle of the allantois, afterwards the urinary bladder.

Fig 11. The lower end of an embryo some hours older than that in Fig. 8. The mucous and vascular layers are drawn upwards, so that not only is the visceral cavity seen, but the lower portion of the intestinal canal, *a*. At the lower portion of the embryo are two small swellings, *b*, *b*, the commencement of the *allantois*. (10 *diam.*)

Fig. 12. The lower end of an embryo twelve hours older than the last. The allantois now forms a sac, the two halves of which, however, are not yet closed. (10 *diam.*)

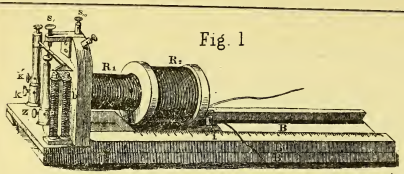
Fig. 10. The embryo of an ovum twelve hours older than the last, suspended by the vascular and mucous layers. All the different parts formerly referred to may be seen further developed. The superior extremity is prominent. In the visceral cavity two long striated bodies are seen, the *Wolffian bodies ;* and the allantois is now so enlarged as to hang out of the visceral cavity, covered with a network of vessels in connection with the vascular layer. (5 *diam.*)

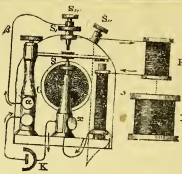
Fig. 14. The head of the same embryo. *a*, Anterior brain-cells—*b*, eyes—*c*, second brain-cell—*d*, first visceral arch—*e*, process thereof—*f*, three lower visceral arches—*g*, right, and *h*, left auricle—*i*, left, and *k*, right ventricle—*l*, aorta, with aortic branches to the visceral arches. (10 *diam.*)

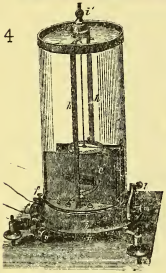
Fig 13. An embryo older than that represented in Fig. 14, seen in front. *a*, Nasal apertures—*b*, eyes—*c*, first visceral arch, now the under jaw—*d*, second visceral arch—*e*, right, and *f*, left auricle—*g*, right, and *h*, left ventricle—*i*, aorta—*k*, liver ; between its two lobes is seen the cut *vena omphalo-mesenterica*—*l*, stomach—*m*, intestinal canal, terminating in the umbilical vesicle—*n*, *o*, Wolffian bodies—*p*, allantois—*q*, upper, and *r*, under extremity. (5 *diam.*)

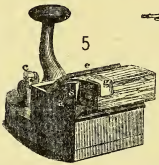
Fig. 15. Embryo of an egg about four weeks old. *a*, Trachea and œsophagus—*b*, thymus gland—*c*, right, and *d*, left auricle—*e*, right, and *f*, left ventricle—*g*, left, and *h*, right aorta —*i*, *i*, *i*, three lobes of the liver—*k*, stomach—*l*, intestinal coils, which by a band, *m* (the former *ductus omphalo-mesentericus*), are in connection with the umbilical vesicle *n*—*o*, Wolffian bodies. (5 *diam.*)

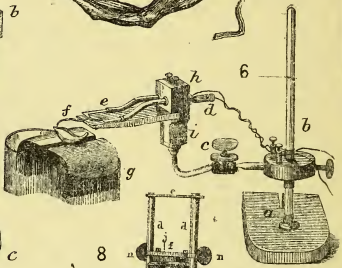
Fig. 1

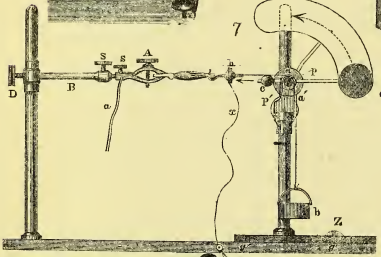


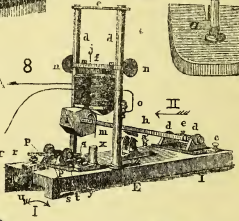


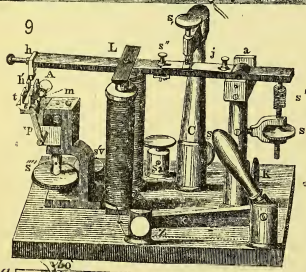


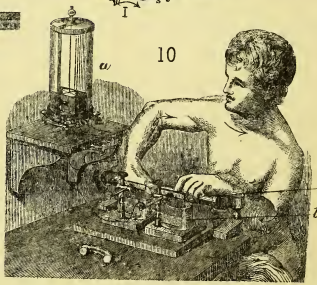












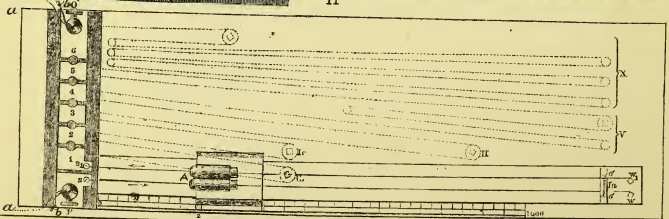


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Plate XIX.—Practical Physiology.

Fig. 1. Du Bois-Reymond's induction-apparatus. R 1. Primary coil; R 2. secondary coil; the upper B is the groove in which the secondary coil slides. B B. Wooden stand. 1. Scale graduated into millimetres. *b*. Electro-magnetic apparatus for attracting Neef's hammer. *k*. Screw for attaching wire from positive pole. Z. Screw for wire in connection with negative pole. k' $S_{///}$. Attachments for a wire when Helmholtz's modification is employed (see Fig. 2 β). S'. Screw, the point of which establishes a connection with the back of the spring.

Fig. 2. The end of Du Bois-Reymond's apparatus, shewing on the right hand a diagrammatic view of the primary and secondary coils. *a*. Screw for attaching the wire β passing to $S_{///}$, as used in Helmholtz's modification. *b*. End of the primary coil. S. Screw point which touches the under surface of the spring, and is placed on the top of the middle pillar, in the base of which there is an attaching screw, *x*. $S_{/}$. Screw, the point of which touches the back of the spring when the apparatus is used in the ordinary way. $S_{//}$. Attachment screw for the wire passing in the direction of the arrow to R_1, the primary coil. The lower arrow indicates the wire passing to the magneto-electric apparatus R_2, secondary coil. *k*. The battery.

Fig. 3. The limb of a frog skinned. *a*. The muscles of the leg; *b*. the sciatic nerve.

Fig. 4. Multiplying galvanometer. *a*. Base; *b*. brass box; C. boxwood frame carrying coils of wire; *f f*. wires leading to galvanometer; *g*. screw for rotating *b*. *h, h*. Vertical brass bars supporting a horizontal bar, from the centre of which the astatic needle is suspended by a single silk fibre. *i'*. Screw for raising or lowering the needle.

Fig. 5. Non-polarizable electrode of Du Bois-Reymond. *a* S. Amalgamated zinc trough; *c*. attachment screws for wires. *e*. A rectangular piece of vulcanite for maintaining the cushion of blotting paper in position; *g*. film of moist clay laid on the cushion so as to protect the muscle from the irritant action of the solution of sulphate of zinc.

Fig. 6. Polarizable electrodes of Du Bois-Reymond. *a*. Wooden stand; *b*. round piece of vulcanite with screws; *c*. universal joint; *d*. binding screws; *h*. square block of ivory with wires, *e*, passing through it; *f*. the nerve lying on the triangular platinum electrodes; *g*. troughs containing cushions of blotting paper immersed in solution of sulphate of zinc.

Fig. 7. Muscle telegraph. A. Forceps holding the femur; B. handle of forceps, bearing at its end the screw S. The forceps may be elongated or shortened by drawing them out of the socket, secured by S. S. Screw for attaching wire *a* from positive pole of the battery; *h*. hook fixed to *tendo Achillis*, and having a wire, *x*, in connection with negative pole attached to it. *a a'* thread passing from *h* over the pulley *p' p* and supporting the bucket *b*; *c*. a round counterpoise-weight attached to the end of the long arm above *p'*, bearing the disc C, which moves in the direction of the arrow. Z. Screw for fixing in the socket the upright pillar of brass bearing the telegraph. *g' g'*. The stand of the whole instrument.

Fig. 8. Pflüger's falling apparatus or trip hammer. E I. Wooden stand; *d d*. uprights bearing the axle *e*, on which the handle *h* of the hammer *i h* moves. *i*. Head of the hammer; *m*. steel point attached to side of the hammer-head, for dipping when the hammer falls, into the trough X. *a' b'* Z. Steel catch for holding securely the handle of the hammer when the head falls. *y*. Screw for attachment of wire in connection with negative pole on the same piece of brass as supports the trough *x*. *c*. Screw for wire from positive pole of battery. P. Lever working between two uprights, P, one end of the lever being seen at S (following the dotted line), and the other at *q*. *r*. Screw point which, when hammer head is elevated, is touched by the end of the lever, but is separated from it when the head falls. *t*. Screw for attachment of wire from positive pole; *u*. screw for the wire in connection with negative pole. The *closing* shock is given by the current passing in the direction *c, d, e, h, m, x, y;* while that of the *opening* shock passes thus: *t, p r, u*. Above the hammer head s the magneto-electric apparatus A for supporting it. *d d*. Two brass upright

pillars ; *c.* transverse bar connecting *d d* at the top. *j.* Bar bearing underneath the two electro-magnets *f.* These may be elevated or depressed on *d d* by the screws *n n*. *O.* Wires connecting two Smee's elements with electro-magnet.

Fig. 9. Heidenhain's tetanometer. K. Upright brass pillar, having an attachment screw, and bearing the lever *h*, L, S″ *j*, *a*. *a*. Fulcrum of the lever ; S″ *j*. two screws by means of which lever may be lengthened or shortened. L. Armature for the electro-magnet seen underneath, S′ V. C. Upright brass pillar bearing at top a horizontal arm, at end of which there is a screw S′, the point of which touches a bit of platinum on the upper surface of the lever. $S_{,,,}$. Attachment screw. $S_{,,}$. Attachment screw. $S_{,,,}$ and $S_{,,,}$ are connected by a wire in the vulcanite stand not shewn. Z. Short brass pillar having a binding screw. The current is broken by pushing back the handle seen on the right, and thus elevating the brass arm *k*. S. Spiral spring, worked by screw S underneath for restraining the action of the lever. A. On left end of the instrument is the apparatus for beating the nerve. *h*. Small ivory hammer, at end of lever ; *h′*. ivory groove in which hammer head *h* beats. *t*. Groove passing transversely for nerve A. *m*. Small roller for attaching the end of the nerve. *p*, Steel spring for retaining roller *m* in position. $S_{,,,,}$. Screw by which the apparatus above may be elevated or depressed.

Fig. 10. Arrangement of apparatus for demonstrating the presence of a current of electricity in the living body. The individual is grasping a wooden roller, having the index fingers immersed in the troughs. On his right hand the galvanometer is seen.

Fig. 11. The rheocord. S W. Platinum wire. S′ $W_{,}$. Another platinum wire. I *a*. Ivory bridge over which the wires pass at *σ σ*. Z. Piece of brass carrying two bottles filled with mercury A, capable of sliding along the platinum wires S W, and S′ $W_{,}$. O. 1000, Scale graduated into millimetres. P S. 1, 2, 3, 4, 5, and 6. Rectangular pieces of brass, which may be connected by brass stoppers, or pegs. P and Q. Short brass pillars, each bearing two attachment screws for wires *a a*. The dotted lines, marked on the right hand thus : $\}$X, $\}$V, II, I *c*, and I *b*, and passing round small ivory pulleys, represent German silver wires, connecting the pieces of brass, 1, 2, 3, 4, 5, and 6, when the stoppers are removed.

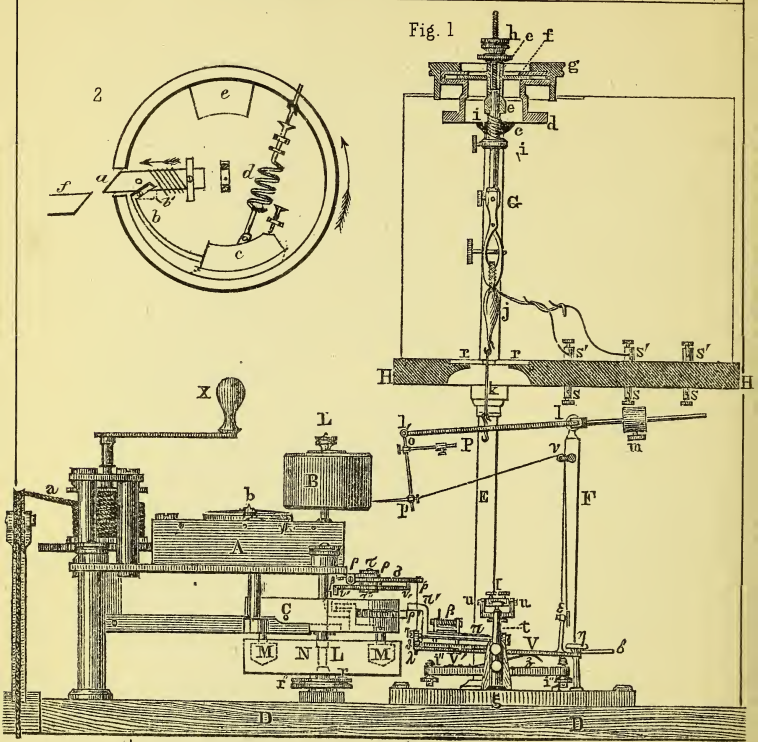
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Plate XX.—Practical Physiology.

Fig. 1. Sectional view of the Myographion. A. Clock-work. B. Revolving cylinder. C. Box containing centrifugal apparatus. D. Wooden stand or base supporting the whole apparatus. E. Strong brass pillar supporting a square glass chamber, in which the nerve preparation is stimulated. F. Brass pillar supporting the lever apparatus *l l*. G. Brass forceps for holding the femur, provided with a binding screw at G. H H. Strong vulcanite floor of the moist chamber, having a round hole between *r* and *r*. L L. Vertical axis on which the cylinder B, and the centrifugal box C, revolve. M N L M is an open box placed beneath the centrifugal box C, but attached to the same axis, L L. In this open box there are two vertical plates, M M, which, by friction with the air, give steadiness to the movement of the machinery. Underneath M N L M, is a screw *r'' r*, for elevating or depressing it. P, on a line with O, a small brass rod, having a movable weight near P, for accurately balancing and adjusting the stylette P, which is observed in contact with the cylinder B. S S S. Three of the four attachment screws for wires coming from Pohl's Commutator. S′ S′ S′. Attachment screws in the interior of the moist chamber, connected through the vulcanite plate H H, with S S S. V′ V. Lever apparatus supporting the mechanism by means of which the primary current is broken in the centre of the bridge.

a. Cord passing from a drum, and having a heavy weight attached to the other end; this is the motive power of the clock-work. The cord may be wound up by the handle X. *b*. Dial for indicating the rapidity of the clock-work. *c*. (Near the top of the figure) a spiral, by means of which the forceps G may be elevated or depressed, and secured by a screw seen opposite. *i, d, i, g*. An apparatus consisting of a circular box in the roof of the moist chamber, in which, by means of an eccentric movement of the plate *f*, and of a vertical movement of the screws *h e*, the forceps G may be brought over exactly the centre of the hole in the vulcanite floor, H H, marked *r k r*. *j*. *Gastrocnemius* muscle, having a hook in the *tendo Achillis*. *k*. Wire passing from the hook just mentioned, to another hook in the lever *l′ l*. *m*. A movable weight on the free end of the lever *l′ l* for balancing the latter. *r r*. Two circular glass plates for almost closing the hole in the vulcanite floor, a small perforation being left for the wire *k*. The more complicated parts of the machinery are marked by Greek letters, and can only be understood by comparing Fig. 1 with Figs. 2 and 3, or by examining the instrument.

Immediately under the point of the stylette P, there may be seen the apparatus for bringing it in contact with the point of the cylinder at the proper moment. ρ ρ ϑ Lever. τ τ″. Axis round which the piece of brass *v′ v′* moves when the end next the axis, L L, is knocked against by a small projection seen close to the axis, and marked β. At *p*, to the right of ϑ is the thread passing downwards to ϑ λ, the end of the lever V′ V. This lever, V′ V moves on an axis in the pillar seen resting on a spring ξ. The spring is attached to a brass bar *i″ i″*, which moves on an axis between two brass pillars, one of which is seen above ξ. On the right of V, there is a delicate pillar of brass, from the top of which, at ε, a thread passes upwards to the pulley *v*, and from thence downwards to the left to the end of the stylette P. Near the bottom of the pillar F, there is a small arm marked *n* δ, by which the lever V′ V may be held down, and the stylette thus withdrawn from the cylinder. Now direct attention to the apparatus for breaking the primary current at a proper moment. In the interior of the centrifugal apparatus C, the dotted outline represents the out-springer, the end of which is seen at ρ ρ. π′ π is the rectangular arm, which, when pushed over to the left by ϱ, the out-springer, breaks the primary current in the centre of the bridge *t*. β. The spring for retaining the arm. Near the base of the pillar E, there is a screw, *u u*, for regulating the action of the lever apparatus V¹ V.

Fig. 2. View of the centrifugal apparatus marked C in Fig. 1. *a*. Point of the out-springer, which moves in the direction of the arrow, propelled by a spiral wound round it. *b*. Steel arm for retaining *a*; attached to a movable weight *c*. The movement of the latter is con-

trolled by a spring, *d*, which may be tightened or relaxed. *e*. Fixed weight acting as a counter-poise to *c*. *f*. End of the rectangular arm seen at *n'*, Fig. 1, and *h*, Fig. 3. The arrow to the right shews the direction in which the box revolves.

Fig. 3. Diagram shewing the arrangement of apparatus for determining the rapidity of the nerve-current. *a*. Forceps holding *femur*; *b. gastrocnemius* muscle; *c. sciatic* nerve; *d*. revolving cylinder; *e*. lever carrying the stylette; *f*. centrifugal apparatus; *g*. steel spring forming the bridge between the two pillars *k i*; *h*. end of out-springer, and above it the end of the rectangular arm; *k*. wire passing from primary coil *m* to one side or pier of the bridge in the myographion; *i*. wire from the other pier back to the battery *l*; *l*. the battery; *m*. primary coil of induction machine; *n*. secondary coil; *n'' n''*. Pohl's Commutator, having at p3, 4 wires going to stimulate a portion of the nerve *c*, close to the muscle at *r*, and at *o* 1, 2, wires passing to *q*, so as to stimulate at a distance from the muscle. The primary circuit is in the direction *l*, *m*, *k*, *g*, *i*, back to *l*. When this is broken at *g*, an induced current is sent from *n* either to *r* or *q*, on the nerve *c*, according as we place the Commutator.

Fig. 4. Du Bois-Reymond's key. B. Vulcanite plate. *b*, *c*. Rectangular pieces of brass, each bearing two screws. These may be connected by means of the handle *d*.

Fig. 5. Pflüger's Myographion. S. Wooden stand. B. Frame having a groove in which the glass plate P moves. F. Brass pillar bearing a square glass chamber for the muscle. L. Forceps for the *femur*, to which is attached the *gastrocnemius* muscle A with the *sciatic* nerve N. *a*. Brass pillars bearing at the top the double lever *b*; *c*. swivel apparatus for connecting the lever *b* with *d*, a long hook attached above to the *tendo Achillis*; *g*. a scale for a weight sufficient to draw down the lever after it has been elevated by the contraction of the muscle; *m*. movable weight for carefully balancing the lever.

Fig. 6. Pohl's Commutator. *a*. A round wooden disc, having six small holes filled with mercury, each having a screw for the attachment of a wire; + *a*. wire coming from positive pole of battery; —*b*. wire going to negative pole; P O. two pieces of wire permanently fixed at one end into + *a*, and — *b*, and the other ends held in close proximity by a glass tube S; *q*, *r*, and *m*, *n*. transverse arcs of wires, having their ends free, so that by moving the bridge P S O, the ends of the arcs may dip into either the holes α β, as seen in the figure, or into A B; α β. attachments for wires going to a nerve or muscle; A B. another pair of the same; *h* and *i*, transverse wires, *h* being bent in the middle so as not to touch *i*. The directions of the current may be as follow: (1.) *Without the transverse wires, h i*. In this case, the current will pass along a circuit, α β, when the ends of the arcs dip into the holes *r n*; but if the bridge be reversed, so that the ends *q*, *r* of the arcs dip into A B, the current will pass along a circuit from A to B. (2.) *With the transverse wires h i*. We can now send a current from α to β, or from β to α, that is, either up or down a nerve. As the bridge is placed in the figure, the direction will be + *a*, P, *r*, α, β, *n*, O,—*b*, and from thence to the battery. But if we now reverse the bridge, so that *q m* dip into the holes A B, the current will travel along the cross wires thus, ——— + α, P, *q*, A, cross wire *h*, *n*, β, α, *r*, cross bar *i*, *m*, O,—*b*, and from thence back to the battery.

Fig. 7. Form of tracings to be obtained on the cylinder of the Myographion. The horizontal line *a b* is the mark made by the stylette during the revolution immediately before the primary current is broken. The two oblique curved lines *c e*, *d f* are produced by the stylette being drawn upwards by the contraction of the muscle when the nerve is stimulated. Of these two, the line *c e* is that produced when the stimulus is applied close to the muscle; while *d f* is caused by the contraction when it is applied at a distance from the muscle. The short distance between the points where these curves leave the horizontal line *c d* indicates the length of time the nerve current occupied in passing from the distant to the near point of stimulation.

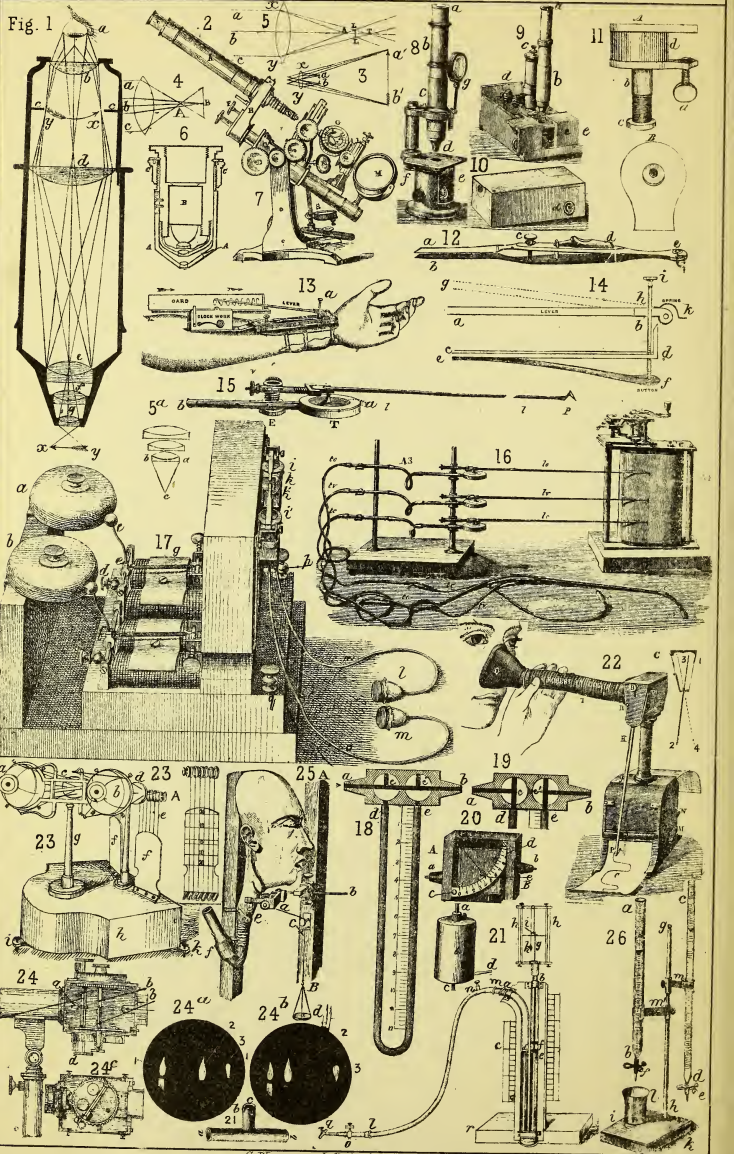

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Plate XXI.—Practical Physiology.

Fig. 1. Section of a compound achromatic microscope. *a.* Eye of observer; *b.* eye-glass; *c c.* stop in the eye-piece; *d.* field glass. The letters, *b, c c,* and *d,* represent the eye-piece, *e, f,* and *g,* the objective, consisting of three achromatic lenses. The arrow, *x, y,* beneath the objective is seen magnified and inverted and curved at *y x.*

Fig. 5. Diagram to illustrate chromatic aberration. *x y.* A bi-convex lens; *b.* a ray of light passing directly through *x y*; *a b.* two rays of light dispersed by *x y,* so that the violet rays come to a focus at A, and the red at T. L L. a screen placed mid-way between A and T.

Fig. 5*a*. View of three achromatic lenses; *a b c* is the angle of aperture of the lenses.

Fig. 3. Diagram to illustrate the theory of enlargement. *x y.* A bi-convex lens through which rays of light pass to the eye from the small object *a b,* and which so refracts these that they enter the eye at such an angle as if they came from a large object *a' b'*. *a b* consequently appears magnified to the size *a' b'*.

Fig. 4. Diagram to illustrate spherical aberration, shewing the rays *a c* impinging on the surface of the lens near the margin brought to a focus at A, while those passing through the centre *b,* not being so much refracted, meet farther off, at B.

Fig. 2. Ross' compound achromatic microscope with movable stage and Gillett's condenser. A. Body of microscope. B. Rectangular arm supporting it. D. Coarse adjustment. F. Fine adjustment. M. Mirror, concave on one side, plane on the other. G. Gillett's condenser fitted beneath the stage. K S. Bull's-eye condensers for reflecting light on opaque objects. 7 is opposite the strong brass pillars supporting the microscope.

Fig. 6. Section of a compound objective of Ross, shewing three achromatic lenses and the arrangement for correcting the lenses for use with covered and uncovered objects. B. Tube carrying the two upper lenses; A A. a cylinder carrying the lower lens; C C. screwed ring for approximating A A to B.

Fig. 8. Oberhauser's microscope. *a.* Eye-piece; *b.* body; *c.* split tube; *d.* objective; *e.* mirror; *f.* fine adjustment; *g.* condenser.

Fig. 9. Nachet's pocket microscope. *a.* Eye-piece; *b.* body; *c.* fine adjustment; *d.* box for containing the microscope and lenses, &c., and on the under surface of the inverted lid the microscope is fixed, as shewn in the figure; *e.* mirror.

Fig. 10. The same instrument seen closed. *d.* A small button for moving the mirror.

Fig. 11. Stirling's section cutting apparatus. A. View from the side. B. View of the top. *a.* Screw for fixing apparatus to a table; *b.* socket in which the fine threaded screw *c* works, pushing up the bottom of the circular box *d.*

Fig. 12. Valentine's knife. a, *b.* Blades; *c.* screw for fixing the distance between the blades; *d.* steel catch for holding blades together at the joint *e.*

Fig. 13. Sphygmograph of Marey affixed to the left wrist. The names describe the parts of the apparatus. The arrows shew the direction in which the card moves by clock-work. *a.* The upright rod in connection with the spring resting on the artery.

Fig. 14. Diagram of the essential parts of the sphygmograph. *a, b.* Lever; *c, d.* fixed bar for attachment of spring *e f*; *f.* button for resting on the pulse; *g h.* dotted lines indicating the position of lever *a b* when elevated; *k.* spring for regulating movements of lever; *i.* head of upright rod, resting below on *f,* and which, by a little metal shoulder, elevates the lever *a b.*

Fig. 15. Marey's drum or tambour for obtaining delicate tracings of pulsations. T. The drum. *a.* Aluminium plate resting on the drum; *b.* tube communicating with the drum. E. Ring by which the apparatus is fixed on an upright brass rod. *l l.* A long light wooden lever. P. A pen point at the end of the lever for making tracings.

Fig. 16. Marey's cardiograph for obtaining simultaneous tracings from different parts of the apparatus. On the right is seen a revolving cylinder for obtaining tracings. *le, lv, lc.* Levers moved by drums, which are seen under A3 connected with india-rubber tubes; *te, tv, tc,* lying in front of the instrument, are india-rubber tubes filled with air, and having at V and *c,* small conical bags, for insertion into the blood vessels or into one of the cavities of the heart.

Fig. 17. Sphygmosphone of Upham, for discriminating between the times of alternate pulsations by sound. *a b.* Bells; *c d.* hammers worked by the two electro-magnets *g*; *e, f.* keepers of the electro magnets *g,* having *c d* attached; *i i'.* bell-shaped glasses, the mouths being covered with india rubber, and having round metallic plates resting on them supporting the levers *k k'*; *l m.* two similar bell-shaped glasses for receiving the impulse from the heart and wrist. The glasses *i i'* and *l m,* and the india rubber-tubing connecting them are filled with water. *p, q.* Connectors for wires leading from a battery and conveying electricity to work the electro-magnets.

Figs. 18 and 19. Volkmann's hæmadromometer for measuring the rapidity of the circulation of the blood. *a b*. Nozzles for insertion into the artery ; *c c*. tubes connected with a stop cock, so that the current may be caused to flow from *a* to *b*, as seen in Fig. 18 or along the U-tube *d e*, as seen in Fig. 19. In Fig. 18 the limb *e* of the U-tube is provided with a scale, but in most instruments the scale passes along the whole length of the U-tube.

Fig. 20. The essential part of Vierordt's hæmatachometer, for measuring the rapidity of the circulation. A B. Square metallic box, two sides being made of glass ; *a b*. nozzles for insertion into the artery ; *c*. a pendulum hanging in the box, near the point of entrance of the blood at *a* ; *d*. a graduated arc for measuring the deviations from the perpendicular of the pendulum ; *e*. the pendulum as moved by a stream of blood through the box.

Fig. 21. The kymographion of Ludwig, for measuring blood pressure, and also for measuring the time occupied by pulsations. *a, d, e*. A U-tube containing mercury, the level of which, in the two limbs, is seen at *d* and *e*. *l, n, m, a*. A tube filled with a solution of carbonate of soda, the part *l, n, m, a*, being made of lead, while *a d* is glass. At *m a* is an accurately fitting screw-collar for uniting the two tubes ; *n*. an air hole in the leaden pipe provided with a stopper ; *l*. a connecting screw-collar between the part of the apparatus *q l*, which is made of brass and the leaden pipe ; *o*. a stop cock ; *g*. a T-shaped nozzle for insertion into the artery. At *c* and *e* are graduated scales opposite each limb of the U-tube, for measuring blood pressure in inches of mercury. The apparatus as described to this point is the hæmadynamometer of Poisseuille. The apparatus for registering the oscillations of the mercury is now added. *f*. A glass float on the surface of the mercury bearing a thin vertical rod *g* ; *b*. a screw-collar ; *h h*. two uprights, having thin wires, on which the transverse bar bearing the stylette *i* moves freely up and down in the same vertical plane ; *k*. a weight which acts as a counterpoise to the float and stylette ; *r*. is the square wooden stand of the instrument. To the left of 21 is seen a revolving cylinder, *b*, moving on an axle, *a c*, and having a stylette, *d*, in contact with it.

Fig. 21*b*. A T-shaped nozzle for insertion into an artery by the ends *a b*, the tube *c* connecting it with the end of the leaden pipe *l n m*, in Fig. 21.

Fig. 22. The anapnograph of Bergeon and Kastus, for measuring the amount of air in inspiration and expiration, and for obtaining a tracing of the movements of respiration. O. India rubber nozzle ; T. india rubber tube ; V. aluminium valve ; H. wooden lever ; P. pen. *c*. Tracing obtained on the paper. N M. Box containing clock-work ; B. button for tightening or relaxing the lever H ; C. diagramatic view of interior of box, shewing 3, 2, the aluminium plate and lever. The dotted line, 4, shews movement of the lever in inspiration.

Fig. 23. The ophthalmotrope of Reute. *a, b*. Models of eye-balls ; *c, d*. brass plates through which cords pass representing the muscles of the eyes ; *e*. cords passing downwards over the brass plates *f, f*. The back of one of these plates *f, f*, graduated, is seen at A. *g*. Brass pillar supporting the apparatus ; *h*. wooden box, having in its interior a transverse roller to which the cords *e*, are attached ; *i, k*. levelling screws.

Fig. 24. The ophthalmometer of Helmholtz. *a, d, b, b*. brass box containing two plates of glass, *b, b*, which may be revolved by the pinion *a, c* ; *d*,. screw-head for moving the pinion. At *c*, is a portion of the telescope. The whole apparatus is mounted on a stand, having a universal joint. 24, *f*. A view from above of a circular brass plate (seen in Fig. 24, in the upper part of the box, in a line with *a*), toothed at the edge, for revolving the glass plates by means of pinions.

Fig. 24*a*. Diagrammatic view of the reflections of a candle flame seen in the human eye, as adjusted for distant objects. 1. Cornea, erect ; 2. anterior surface of lens, erect ; 3. posterior surface of lens, inverted.

Fig 24*b*. The same as adjusted for near objects. The anterior surface of the lens has become more convex, as 2 is seen nearer to 1 than in 24*a*.

Fig. 25. Müller's apparatus for shewing the production of voice. *a*. Forceps for compressing the thyroid cartilage, so as to approximate the cords ; *b*. movable handle of the forceps ; *c*. cord passing from a hook attached to the upper margin of the thyroid cartilage, in the mesial line, over a small pulley ; *d*. scale or balance, at the end of *c*. By placing weights in *d*, the tension of the vocal cords may be increased at pleasure. A B. Wooden pillar supporting the forceps and pulley.

Fig. 26. A stand, *i k*, on which there is a pillar, *g h*, bearing two of Mohr's burettes, which may be elevated or depressed by the split tubes *m m* ; *a, b*, and *c, d*. glass burettes, graduated in millimetres ; *f, e*. Mohr's clips for compressing the short pieces of india-rubber tubing ; *l*. glass beaker placed under *a, b*.

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